PATENT SPECIFICATION

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(19)

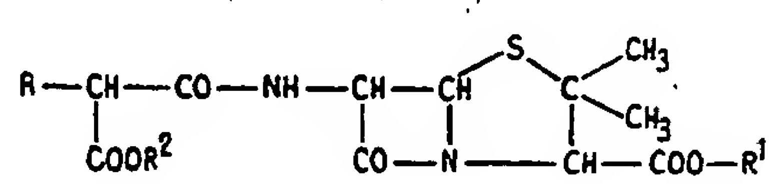
(72) Inventors BERTIL AKE EKSTROM, ODON KALMAN JOZSEF KOVACS and BERNDT OLOF HARALD SJOBERG

(54) PENICILLINS

(71) We, ASTRA LAKEMEDEL, AKTIEBOLAG, a Swedish Body Corporate of Kvarnbergagatan 16 S-151 85 Sodertalje, Sweden, do hereby declare the invention for which we pray that a patent may be granted to us, and the method by which it is to be performed, to be particularly described in and by the following statement:—

The present invention relates to new penicillins, to methods for their preparation, to pharmaceutical preparations containing them and to the use of the penicillins in combating infection.

The present invention provides penicillins of the general formula



I

and pharmaceutically acceptable salts thereof, in which

R is phenyl, thienyl or furyl group and

R^1 is hydrogen or a

$$-\underset{}{\overset{R^3}{\overset{|}{C}H}}-O-\overset{O}{\overset{\|}{C}}-O-R^4$$ group and

R^2 is a

$$-\overset{R^3}{\overset{|}{C}H}-O-\overset{O}{\overset{\|}{C}}-O-R^4$$

group or hydrogen or an alkyl group of 1 to 8 carbon atoms, an aryl group or an aralkyl group,

R^3 is hydrogen or a methyl group;

R^4 is an alkyl, alkenyl or alkynyl group of up to 8 carbon atoms, a cycloalkyl group of 3 to 7 carbon atoms or a phenyl benzyl, indanyl, thienyl, furyl, furfuryl, pyridyl, pyridylmethyl or 2-methyl-1,3-dioxanyl group, the said groups being unsubstituted or substituted with one or more amino, substituted amino, halogeno or nitro radicals;

provided that R^2 is

$$-\overset{R^3}{\overset{|}{C}H}-O-\overset{O}{\overset{\|}{C}}-O-R^4$$ when R^1 is a hydrogen atom.

Illustrative examples of radicals included in the above definitions are:

alkyl: methyl, ethyl, propyl, isopropyl, butyl, isobutyl, pentyl, hexyl, heptyl, octyl, 2-ethyl-hexyl;

cycloalkyl: cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl;

alkoxy: methoxy, ethoxy, propyloxy, isopropyloxy, butoxy, isobutoxy;

halogen: F, Cl, Br;

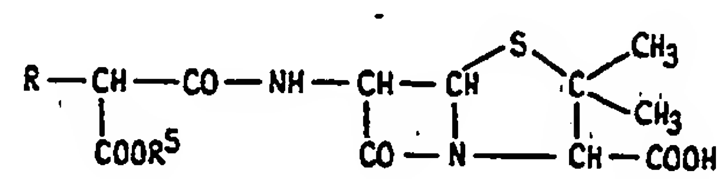
aryl: phenyl, naphthyl, 5-indanyl;

aralkyl: benzyl, naphthylmethyl.

The compounds of the invention are of value in the treatment of infectious diseases in man or animal caused by bacterial organisms. They may be isolated and used as such but also, depending on the presence of basic or acidic groups in the molecule, in the form of salts with pharmaceutically acceptable organic or inorganic acids or bases. Examples of suitable acids are hydrochloric acid, hydrobromic acid, sulphuric acid, phosphoric acid, acetic acid, tartaric acid, citric acid, and fumaric acid. Examples of suitable bases are sodium hydroxide, potassium hydroxide, calcium hydroxide, aluminium hydroxide, ammonium hydroxide, non-toxic amines as trialkylamines, including triethylamine, procaine, dibenzylamine, N-benzylbetaphenethylamine, 1-ephenamine, N,N'-dibenzyl-ethylenediamine, dehydroabietylamine, N,N'-bis-dehydroabietylethylene-diamine, N-(lower)-alkyl-piperidine (e.g. N-ethyl-piperidine) and other bases which have been used for the preparation of salts with penicillins.

The side chain of the penicillin structure in formula I contains an asymmetric carbon atom in the α-position. Depending on the configuration around this carbon atom the compounds will occur in two different diastereoisomeric forms which are both biologically active. Likewise the ester groups may contain asymmetric atoms, e.g. when $R^3 = CH_3$, giving rise to different diastereoisomeric forms which also all are biologically active. It is to be understood that the invention comprises the pure diastereoisomers as well as mixtures of them and the process for preparing them by resolving a mixture of stereoisomers obtained by one of the processes described below.

It is known that substitution of benzylpencillin and analogous compounds in the α-position of the side chain with a carboxy group or certain esterified carboxy groups gives compounds of the general structure:



II

where R has the same meaning as above and R^5 is hydrogen, alkyl or aralkyl groups, which show good antibacterial activity against grampositive and gramnegative bacteria, including *Pseudomonas aeruginosa* (Neth. patent specification 6 404 384, South African patent specification 67/2804, South African patent specification 67/6472, Neth. patent specification 6 805 524, U.S. patent specification 3 142 673, Neth. patent specification 6 913 416).

Such compounds are, however, poorly or only moderately absorbed when administered orally, and the carboxy compounds (II, $R^5 = H$) have to be given by injection. It is one purpose of the present invention to provide esters of these compounds which are well absorbed orally and then hydrolysed within the body to give blood and organ levels of the compounds of the general formula II that are adequate for the treatment of infectious diseases, caused by bacteria sensitive to penicillins of the general formula II.

The carboxy groups of the α-carboxypenicillins (II, $R^5 = H$) is rather unstable and is partly split off during the preparation of the compounds and on storage to give the corresponding non-carboxylated penicillins which are less active against the gram-negative bacteria, and especially so against *Ps. aeruginosa*. By transforming the carboxy group into an ester group this decomposition is avoided and compounds are obtained which are readily prepared and stored. To achieve the full antibacterial activity of the α-carboxypenicillin it is, however, necessary to choose such ester groups that are rapidly hydrolysed *in vivo* to release the carboxy penicillin. The present invention to provides esters which are stable during production and under storage conditions but which, after absorption in the organism, are rapidly hydrolysed to give high blood and organ levels of carboxy-penicillins.

The compounds of formula I are well tolerated, give rise to a low frequency of side effects and may readily be used in pharmaceutical compositions, either as

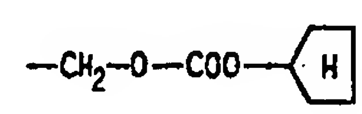
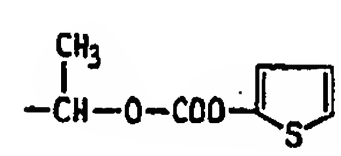
such or in the form of their salts, and they can be intermixed with solid carriers or adjuvants or both. In such compositions the ratio between the therapeutic substance and the carriers and adjuvants may vary between 1% and 95%. The compositions may either be formulated for instance, as tablets, pills or dragees or can be supplied in containers, such as capsules or as mixtures they may be bottled. Pharmaceutically acceptable, organic or inorganic, solid or liquid carriers may be used, suitably for oral or enteral administration or for topical application, in manufacturing the compositions. Gelatine, lactose, starch, magnesium stearate, talc, vegetable and animal fats and oils, natural rubber and polyalkylene glycol and other known carriers for pharmaceuticals are all suitable for manufacturing compositions of said compounds. The preferred salt of the compounds of the invention is the hydrochloride, but salts with other inorganic or organic acids, also antibiotically active acids, may be used, for instance phosphates, acetates or salts with phenoxymethylpenicillin. Moreover the compositions may contain other pharmaceutical active components, suitable for administration with the compound of the invention when treating infectious diseases, for instance, other suitable antibiotic substances, e.g. gentamycin and polymyxin.

In the treatment of bacterial infections in man, the compounds of invention are for example administered in amounts corresponding to 5 to 200 mg/kg/day, preferably in the range of 10 to 100 mg/kg/day in divided dosages, e.g. two, three or four times a day. They are administered in dosage units containing e.g. 175, 350, 500 and 1000 mg of the compounds.

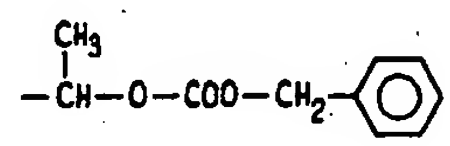
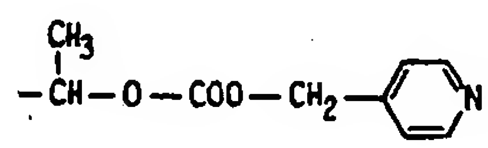
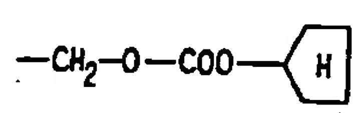
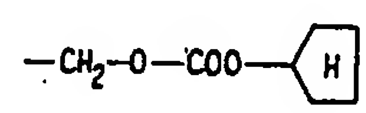
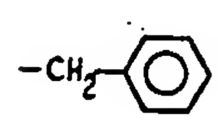
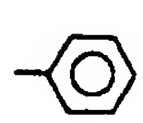
The invention includes within it scope a method of combatting infections in animals excluding man which comprises administering to the animal a compound of formula I or a salt thereof, or a composition containing said compound or salt.

Examples of preferrred compounds of the invention are:

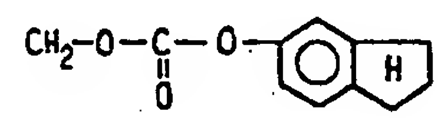
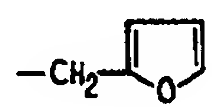
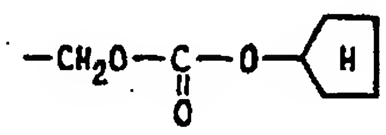
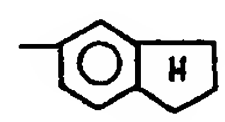
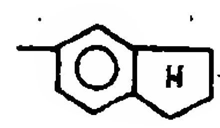
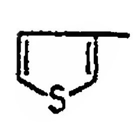
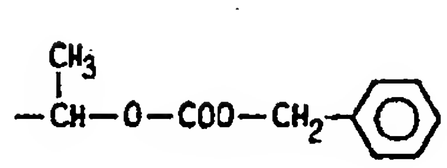
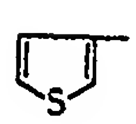
$$R-\underset{\displaystyle COOR^2}{CH}-CONH-APA-COOR^1$$

R	R^2	R^1
C_6H_5	H	$-CH(CH_3)-O-C(=O)-O-CH_2-C(CH_3)_2-CH_3$
C_6H_5	H	$-CH(CH_3)-O-C-COO-C_2H_5$
C_6H_5	H	$-CH_2-O-COO-C_2H_5$
C_6H_5	H	
C_6H_5	H	$-CH(CH_3)-O-COO-CH_2-CH_2-NH-CH_3$
C_6H_5	H	$-CH(CH_3)-O-COO-CH_2-CH_2-NH-C(=O)-CH_3$
C_6H_5	H	$-CH_2-O-COO-CH_2-CH_2-NH-C(=O)-CH_2Cl$
C_6H_5	H	$-CH_2-O-COO-CH_2-CH_2-NH-C(=S)-CH_3$
C_6H_5	H	
C_6H_5	$-CH_2-O-COO-CH_3$	H

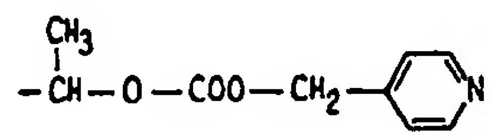
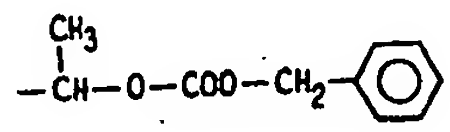
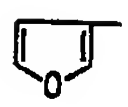
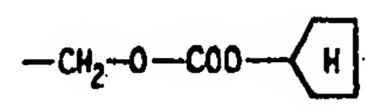
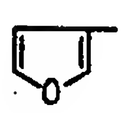
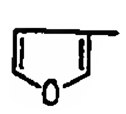
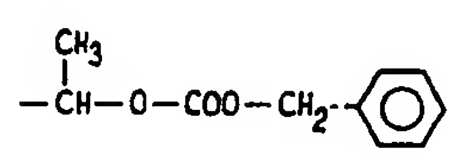
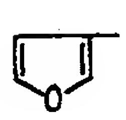
$$R-\underset{\substack{|\\ COOR^2}}{CH}-CONH-APA-COOR^1$$

R	R²	R¹
C_6H_5	$-CH_2-O-COO-CH_2-CH_2-\overset{\substack{Me\\ \vert}}{\underset{\substack{\vert\\ Me}}{CH}}-Me$	H
C_6H_5		H
C_6H_5		H
C_6H_5	$-\overset{\substack{CH_3\\ \vert}}{CH}-O-COO-C_2H_5$	$-\overset{\substack{CH_3\\ \vert}}{CH}-O-COO-C_2H_5$
C_6H_5	$-CH_2\cdot O-COO-C_2H_5$	$-CH_2-O-COO-C_2H_5$
C_6H_5		
C_6H_5	$-\overset{\substack{CH_3\\ \vert}}{CH}-O-COO-C_2H_5$	$-\overset{\substack{CH_3\\ \vert}}{CH}-O-COO-CH_2-CH=CH_2$
C_6H_5		$-\overset{\substack{CH_3\\ \vert}}{CH}-O-\underset{\substack{\Vert\\ O}}{C}-O-CH_2CH_2NH_2$
C_6H_5		$-\overset{\substack{CH_3\\ \vert}}{CH}-O-\underset{\substack{\Vert\\ O}}{C}-O-C_2H_5$

$$R-\underset{\displaystyle COOR^2}{CH}-CONH-APA-COOR^1$$

R	R²	R¹
C_6H_5	$-C(CH_3)_3$	
C_6H_5		
C_6H_5		$-\underset{\displaystyle CH_3}{CH}-O-\underset{\displaystyle \overset{\|}{O}}{C}-O-C_2H_5$
C_6H_5		$-CH_2-O-\underset{\displaystyle C}{C}-O-C_2H_5$
	H	$-CH_2-O-COO-CH_2-CH_2-NH-\overset{\displaystyle S}{\overset{\|}{C}}-CH_3$
		H
	$-\underset{\displaystyle CH_3}{CH}-O-\underset{\displaystyle O}{C}OC_2H_5$	$-\underset{\displaystyle CH_3}{CH}-O-\underset{\displaystyle O}{C}OC_2H_5$
	H	$-\underset{\displaystyle CH_3}{CH}-O-COO-CH_2-CH=CH_2$

$$\begin{array}{l} R - CH - CONH - APA - COOR^1 \\ \quad\;\; | \\ \quad\; COOR^2 \end{array}$$

R	R^2	R^1
		H
		H
	H	
	$-\underset{}{\overset{CH_3}{\overset{\vert}{CH}}} - O - \underset{\underset{O}{\Vert}}{C} - OC_2H_5$	$-\overset{CH_3}{\overset{\vert}{CH}} - O - \underset{\underset{O}{\Vert}}{C} - OC_2H_5$
	H	$-CH_2-O-COO-CH_2-CH_2-NH-\overset{\overset{O}{\Vert}}{C}-CH_2Cl$
		H
	$-\overset{CH_3}{\overset{\vert}{CH}}-O-COO-CH_2-CH_2-NH_2$	$-\overset{CH_3}{\overset{\vert}{CH}}-O-COO-CH_2-CH_2-NH_2$

Preferred classes of compounds of the invention are such compounds of formula I, where R is phenyl, 2- or 3-thienyl or 2- or 3-furyl;

R^2 is hydrogen, lower alkyl, benzyl, phenyl, 5-indanyl, lower alkoxycarbonyloxymethyl, 1′-lower alkoxycarbonyloxyethyl, phenoxycarbonyloxymethyl, 5-indanyloxycarbonyloxymethyl, 1′-phenoxycarbonyloxy-ethyl, or 1′-(5-indanyloxy)carbonyloxy-ethyl, and

R^1 is lower alkoxycarbonyloxymethyl, 1'-lower alkoxycarbonyloxy-ethyl, phenoxycarbonyloxy-methyl, 5-indanyloxycarbonyloxy-methyl, 1'-phenoxy-carbonyloxy-ethyl, or 1'-(5-indanyloxy)carbonyloxy-ethyl.

Further classes of preferred compounds of the invention are those in which R^1 is hydrogen and R^2 is lower alkoxycarbonyloxymethyl, 1'-lower alkoxycarbonyl-oxy-ethyl, phenoxycarbonyloxy-methyl, 5-indanyloxycarbonyloxy-methyl, 1'-phenoxycarbonyloxy-ethyl, 1'-(5-indanyloxy)carbonyloxy-ethyl.

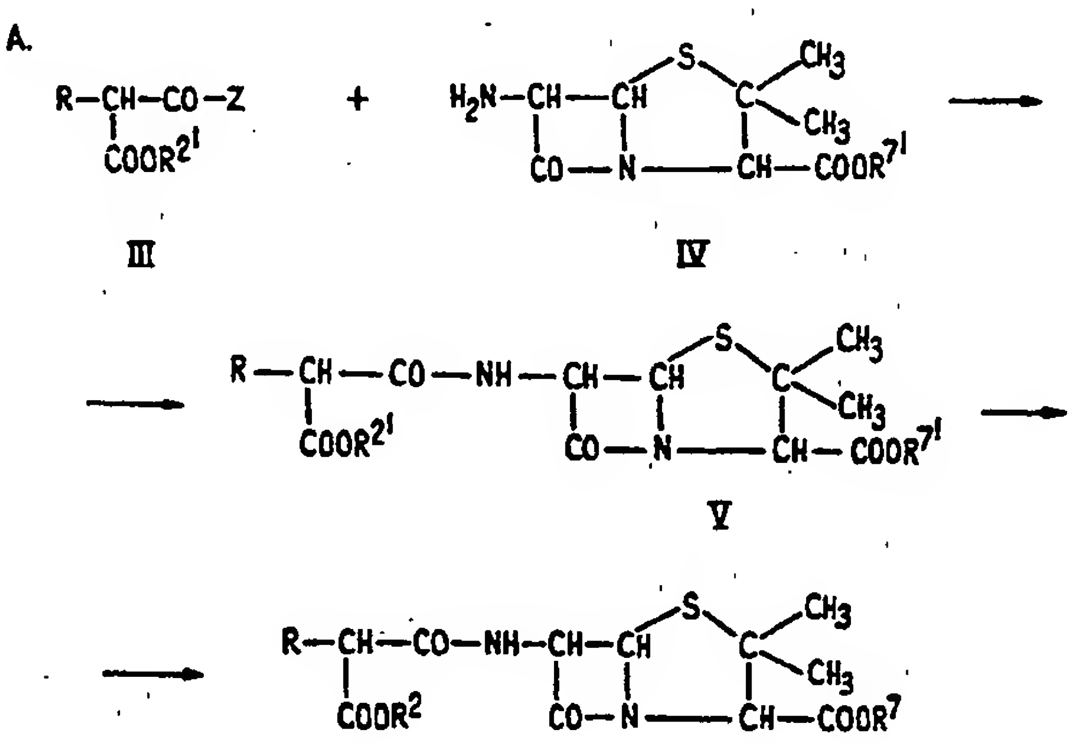
("Alkyl" and "alkoxy" radicals referred to herein as "lower" contain a maximum of 8 carbon atoms.

The alkoxycarbonyloxy groups in R^1 and/or R^2 may be substituted by amino, methylamino or dialkylamino groups.

The compounds of the invention may be prepared in different ways, as follows:

Preparation of esters of the penicillins

A.



IA

According to this method an activated malonic ester derivative III is reacted with an ester of 6-aminopenicillanic acid (6-APA) IV to form a pencillin ester V.

When $R^{2'} = R^2$ and $R^{7'} = R^7$ the product V is a compound of the invention. When $R^{2'}$ or $R^{7'}$ contain groups that are protected, the protecting groups are removed in *per se* known manner in at least one additional step to give the compounds of the general formula I A.

In the formula scheme above the different radicals have the following definitions:

R has the previously given definition,

$R^{2'}$ is R^2, as defined above, or when R^2 is a hydrogen atom or when R^2 contains amino or substituted amino groups, a protected derivative of R^2,

—CO—Z is a reactive group capable of reacting with an amino group to form an amide, e.g. an acid chloride or its functional equivalent.

$R^{7'}$ is R^7, or when R^7 contains amino or substituted amino groups, a protected derivative of R^7,

R^7 is

$$-\overset{R^3}{\overset{|}{C}}H-O-\overset{O}{\overset{\|}{C}}-O-R^4$$

R^3 and R^4 have the meaning given previously.

As protecting groups for the carboxyl group, groups that have been used as carboxylprotecting groups in the penicillin synthesis may be used. More particularly, the protecting group may be benzyl, p-nitro-benzyl or diphenylmethyl, which groups can be split off by catalytic hydrogenation or the protecting group may be an alkyl or an acyl group that can be removed by mild alkaline hydrolysis, or the protecting groups may be a β-trichloroethyl group that can be removed by treatment with zinc in acetic acid, or the protecting group may be a β-iodoethyl, an α-p-tolylsulphonylethyl or a mono- or dihalogenobenzyl group which can be removed by treatment with basic agent, e.g. sodium thiophenolate.

The protecting groups for the amino and the substituted amino groups must be such that they can be removed without destruction of the penicillin ring system. Such protecting groups known to the art are e.g. the benzyloxy carbonyl, the o-nitrophenylsulphenyl, the 2-p-tolylsulphonyl-ethoxy-carbonyl, the β-trichloroethoxy-carbonyl and the 1-methoxycarbonylpropen-2-yl group.

The reaction is an acylation of an ester of 6-aminopenicillanic acid and can be performed in the manner described for acylation of other esters of 6-aminopenicillanic acid (e.g. as described in the French patent specification 1 567 027). The acylating group —CO—Z in III may be an acid chloride group, or a group functioning in the same way, e.g. an acid bromide, an acid azide, an anhydride, a mixed anhydride formed with an inorganic acid or an organic acid such as an alkylcarbonic acid, for instance isobutyl carbonic acid, a carbonic acid, a sulphonic acid and especially an alkoxyformic acid or may be a radical obtained by reacting the α-substituted phenylacetic acid and a carbodiimide or N,N^1-carbonyldiimidazol or another compound reacting in a similar way.

The reaction can be performed in organic solvents such as diethyl ether, tetrahydrofuran, acetone, ethyl acetate, chloroform, methylene chloride, dimethylformamide, dimethyl sulphoxide, or hexamethylphosphoramide, in water or in aqueous organic solvents in presence of organic or inorganic bases such as triethylamine, quinoline, pyridine, N-methyl-morpholine, sodium hydroxide, sodium bicarbonate or potassium carbonate.

The compound of the general formula V may be isolated by extraction from the reaction mixture, if necessary after dilution with water and neutralization.

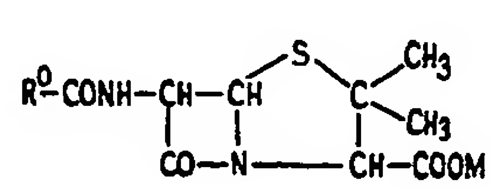
The compounds of the general formula V ($R^{2'} = R^2$; $R^{7'} = R^7$) are compounds of the invention of the general formula I.

The esters of 6-aminopenicillanic acid with the general structure IV may be prepared by treatment of 6-APA with compounds $R^{7'}$ — Y, where $R^{7'}$ has the same meaning as above and Y is halogen or a functionally equivalent derivative thereof such as an organic sulphonic acid residue. The reaction is preferably performed in organic solvents like dimethylformamide or dimethylsulphoxide.

Alternatively 6-acylaminopenicillanic acids with acyl groups that can be removed without destruction of the penicillin ring system are treated with $R^{7'}$ — Y to give esters of the 6-acylaminopenicillanic acids from which the acyl groups then are removed to give the esters of 6-aminopenicillanic acid of the formula IV. A preferred method consists of reacting a salt, e.g. the sodium, potassium or tetra-alkylammonium salt of benzylpenicillin with $R^{7'}$—Y, in an organic solvent like dimethylformamide, dimethylsulphoxide, acetone, chloroform, methylene chloride or hexamethylphosphoramide or in a mixture of an organic solvent and water, e.g. aqueous acetone or dioxane to give the corresponding ester of benzylpenicillin. The phenylacetyl side chain is then removed according to the method described in Neth. patent specification 6 401 421 or South African patent specification 67/2927 by treatment with phosphorus pentachloride in presence of a tertiary organic base to give an imino chloride which is reacted with an alcohol such as propanol, to give the corresponding imino ether which is hydrolysed by addition or water or alcoholized by addition of alcohol to give the ester IV. Alternatively the phenylacetyl side chain may be removed by enzymatic hydrolysis using an *E. coli* acylase according to the method described in French patent specification 1 576 027.

In still another method N-protected 6-aminopenicillanic acids are reacted with $R^{7'}$—Y, where $R^{7'}$ and Y are as defined above to give the corresponding ester from which the protecting groups are removed to give the compounds of the general formula IV. Examples of protecting groups which can be used are the benzyloxycarbonyl group which is removed by catalytic hydrogenation, the o-nitro-phenylsulphenyl group which can be removed by treatment with nucleophilic agents at acid pH (Japanese patent specification 505 176) and the trityl group which can be removed by mild acid hydrolysis.

B. A natural or biosynthetic penicillin of the formula



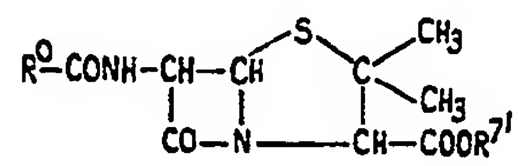
VI

where R°CO represents the acyl group in the side chain of the natural or biosynthetic penicillin and M represents hydrogen or an alkali metal atom such as

sodium or potassium, is esterified by reaction with a compound of the formula

$$R^{7'} — Y \qquad \text{VII}$$

where $R^{7'}$ and Y have the meanings specified above, whereafter the ester of the formula

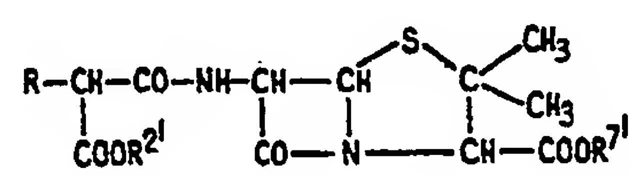


VIII

thus formed where R°CO and $R^{7'}$ are as defined above is reacted with a phosphorus halide in an inert solvent and suitably in presence of a tertiary amine to give an imino halide compound, which is reacted with a lower alcohol to give an iminoether derivative, which imino ether thereafter is reacted with a compound of the formula

$$\begin{array}{l} R—CH—CO—Z \\ \quad\;\; | \\ \quad COOR^{2'} \end{array} \qquad \text{III}$$

wherein R, $R^{2'}$ and Z have the meanings specified above, and the reaction product treated with water or an alcohol to give a compound of the formula

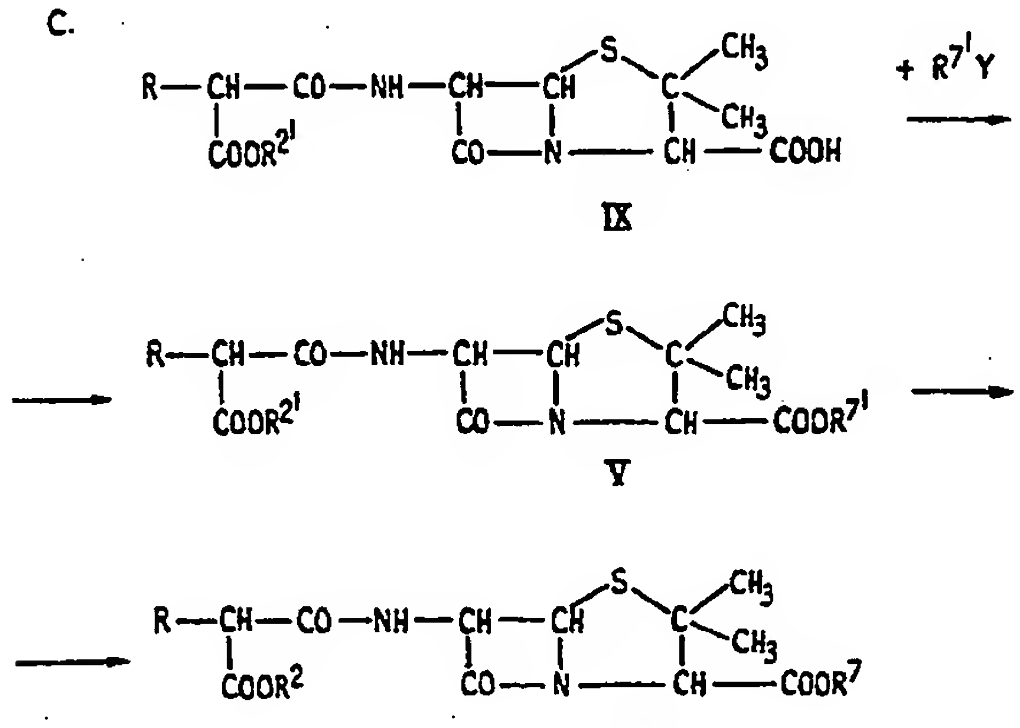


V

wherein R, $R^{2'}$ and $R^{7'}$ are as defined above which compound is then converted to a compound of the formula I as described under A above. In this method the intermediate imino ether compound is directly acylated without isolation of any intermediate products.

The groups R°CO— in the compound of the formula VI is an organic acyl group contained in known natural or biosynthetic penicillins. Thus the groups R° may be an alkyl, aralkyl or a methyl substituted with a heterocyclic group and derivatives thereof. Examples of suitable groups R° are heptyl, phenoxymethyl, 2-thienylmethyl, 2-furylmethyl, and benzyl. Examples of suitable phosphorus halides are phosphorus pentachloride, phosphorus pentabromide, phosphorus oxychloride, and phosphorus trichloride. Phosphorus pentachloride is preferred. Examples of suitable alcohols with which the imino halide may be treated are lower alkyl alcohols such as methanol, ethanol, and n-propanol.

C.



IX → V → IA

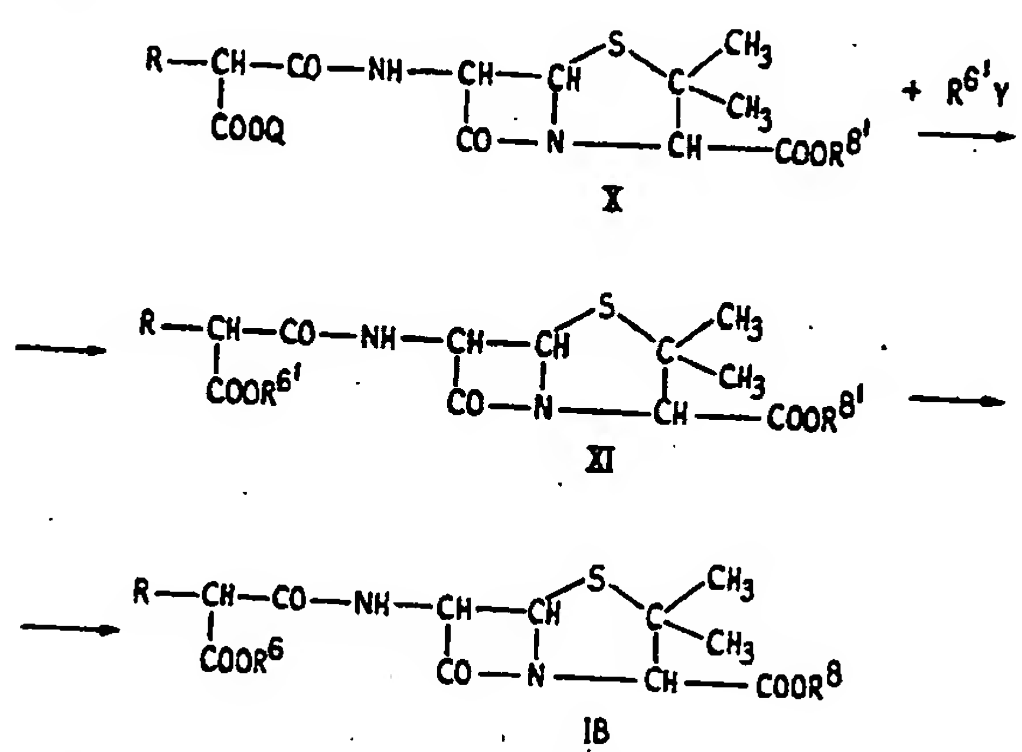
The compounds of the general formula IX wherein R and $R^{2'}$ are as defined above, may, in the form of a salt, be converted into a compound of general formula

V by reaction with compounds of the formula $R^{7'}Y$ where $R^{7'}$ has the same meaning as above and Y is halogen or a functionally equivalent group thereof such as a sulphonyl acid residue. When Y is halogen, preferably chlorine, bromine or iodine, or when it is a sulphonic acid residue, e.g. a p-tolyl-sulphonyloxy group, the reaction is preferably performed with a salt, e.g. sodium, potassium, trialkylammonium or tetraalkyammonium salt of the compound IX in an organic solvent such as dimethylformamide, dimethylsulphoxide, acetone, chloroform or methylene chloride or in a mixture of water and an organic solvent, e.g. aqueous dioxane or acetone.

When $R^{2'} = R^2$ and $R^{7'} = R^7$ the product V is a compound of the invention. When $R^{2'}$ or $R^{7'}$ contain groups that are protected, the protecting groups are removed in *per se* known manner in at least one additional step to give the compounds of the general formula I A.

Penicillins with the formula IX, where $R^{2'}$ is as defined above, are prepared by acylating 5-aminopenicillanic acid according to methods known to the art.

D.



In formula X Q is H or C cation. The compound of the formula X may be reacted in the form of a salt with a compound of the formula $R^{6'}Y$ to form a compound of the formula XI. $R^{6'} = R^6$ and $R^{8'} = R^8$ the product XI is a compound of the invention. When $R^{6'}$ or $R^{8'}$ contain groups that are protected, the protecting groups are removed in *per se* known manner in at least one additional step to give the compounds of the general formula I B.

In the formula scheme above the the different radicals have the following definitions:

R has the previously given definition,

$R^{6'}$ is R^6 or when R^6 contains amino or substituted amino groups, a protected derivative of R^6,

R^6 is

$$-\underset{\displaystyle R^3}{\overset{|}{C}}\!H-O-\overset{\displaystyle O}{\overset{\|}{C}}-O-R^4,$$

an alkyl group containing from 1 to 8 carbon atoms, an aryl group or an aralkyl group,

$R^{8'}$ is R^8 or when R^8 contains amino or substituted amino groups, a protected derivative of R^8,

R^8 is

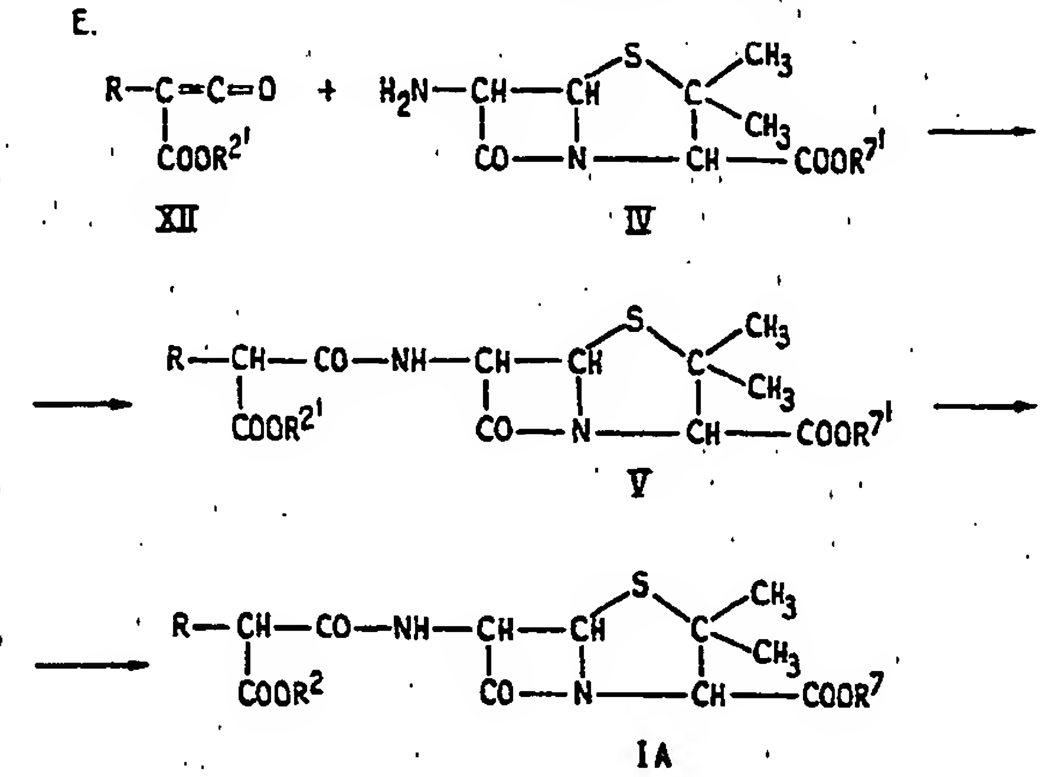
$$-\underset{\displaystyle R^3}{\overset{|}{C}}\!H-O-\overset{\displaystyle O}{\overset{\|}{C}}-O-R^4$$

R^3, R^4 and Y have the meaning given previously.

As protecting groups, the protecting groups mentioned under A are applicable.

The reaction conditions described under C are, in applicable parts, also valid for this method.

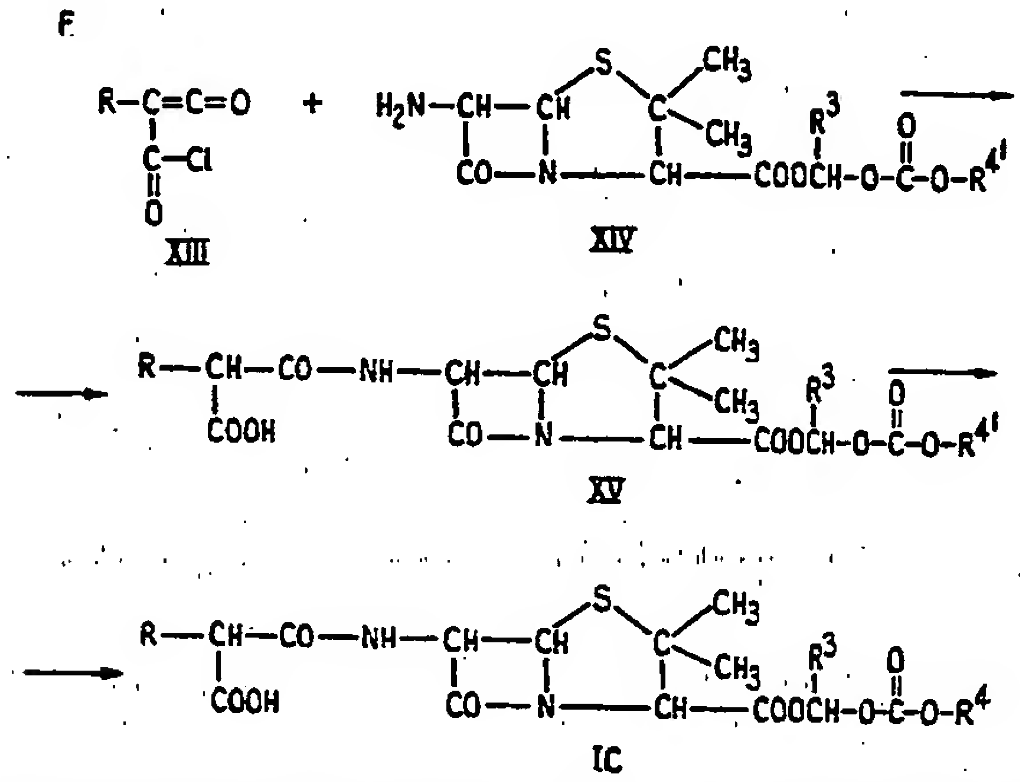
E.



where R, R^2, $R^{2'}$, R^7 and $R^{7'}$ are as defined above.

Instead of the activated carboxylic acid derivative with formula III used in method A a ketene derivative of the formula XII may be used in the acylation reaction as described in Belgian patent specification 726 421.

F

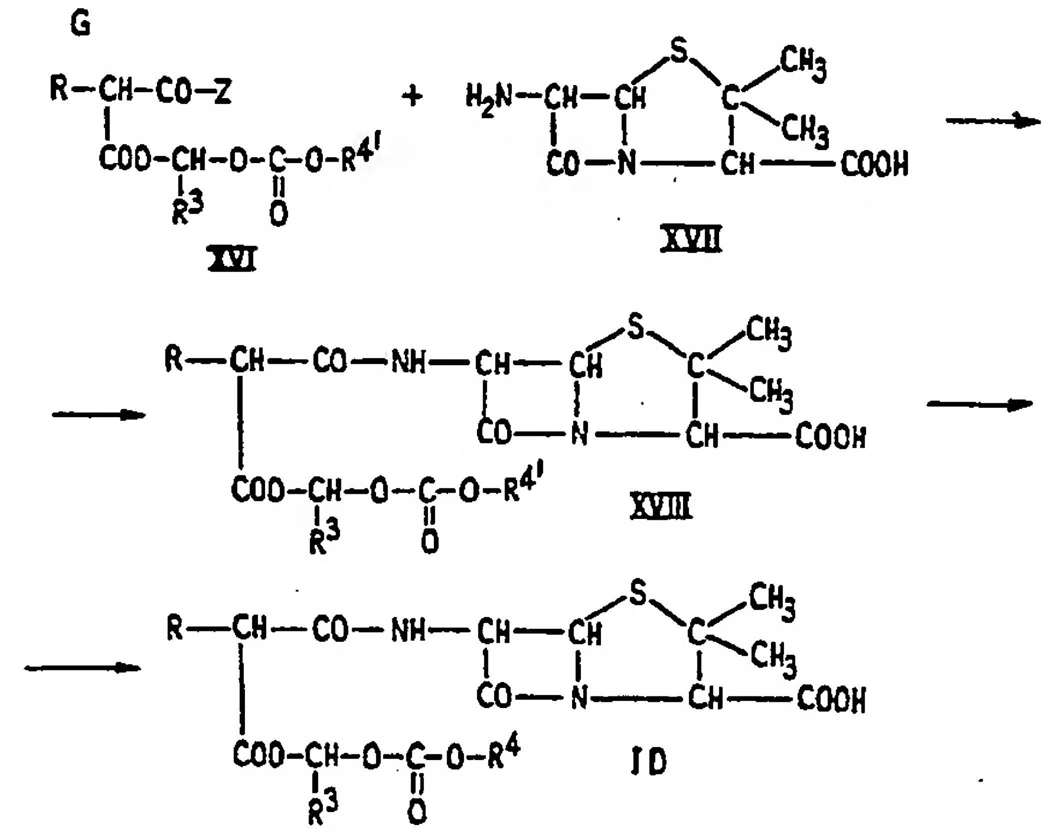


A ketene acid chloride of the formula XIII is reacted with the 6—APA—ester of formula XIV whereafter the obtained compound is hydrolysed to form a compound of the formula XV. When $R^{4'}=R^4$ the product XV is a compound of the invention. When $R^{4'}$ contains groups that are protected, the protecting groups are removed *per se* known manner in at least one additional step to give the compounds of the general formula I C.

In the formula scheme above the radicals R, R^3, and R^4 have the previously given definition and $R^{4'}$ is R^4 or when R^4 contains amino or substituted amino groups, a protected derivative of R^4.

As protecting groups, the protecting groups mentioned under A are applicable.

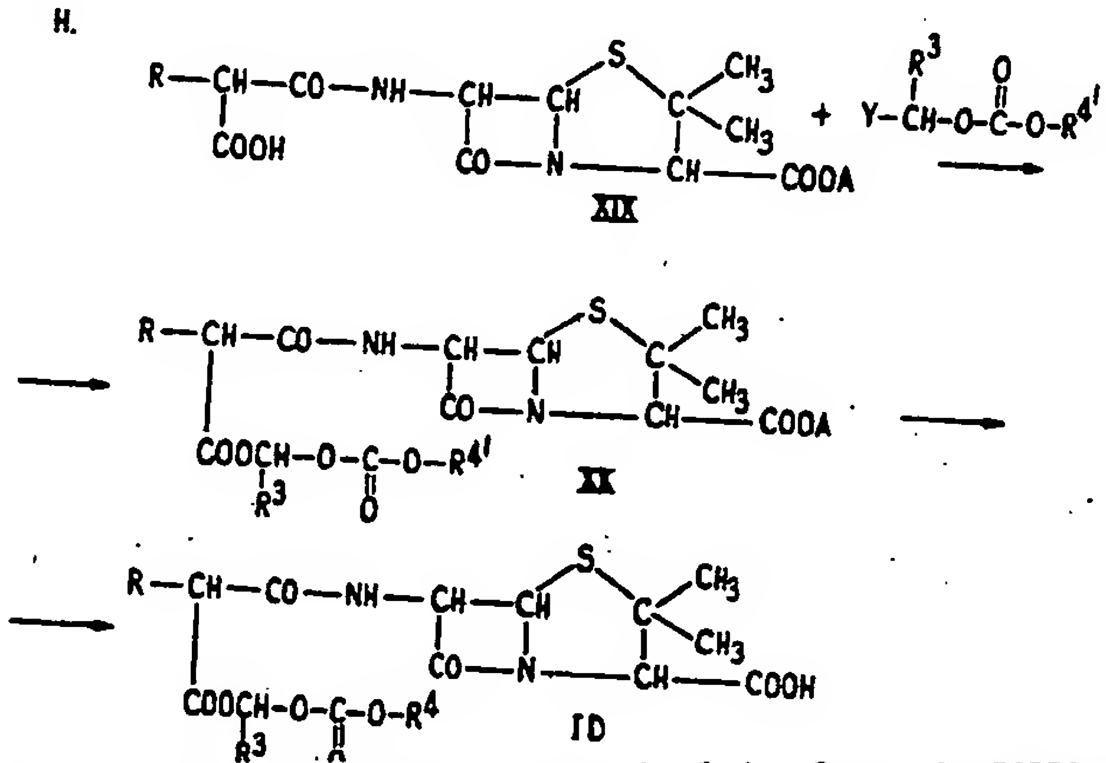
G



According to this method an activated malonic ester derivative XVI is reacted with 6-aminopenicillanic acid (6—APA) XVII to form a penicillin of the formula XVIII, in which formulas R, —CO—Z, R^3, $R^{4'}$, and R^4 are as defined above.

The reaction conditions are these which can be used for the preparation of penicillins by acylation of 6—APA. The conditions described for the preparation of the corresponding esters (method A) may in applicable parts also be valid for this method.

H.



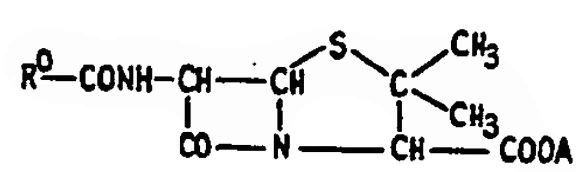
According to this method a compound of the formula XIX wherein R is as defined above and A is a protecting group, is reacted with a compound of the formula

$$Y{-}\overset{\displaystyle R^3}{\overset{|}{C}H}{-}X{-}\overset{\displaystyle O}{\overset{\|}{C}}{-}O{-}R^{4'}$$

wherein Y, R^3, R^4, and $R^{4'}$ are as defined above to form a compound of the formula XX, which compound is then converted to a compound of the formula I D by replacing the groups A with a hydrogen atom, and by replacing the protecting groups of $R^{4'}$ in *per se* known manner.

As A, groups that have been used as carboxylprotecting groups in penicillin synthesis may be used. Especially A may be benzyl, p-nitro-benzyl or diphenylmethyl, which groups can be split off by catalytic hydrogenation or A may be an alkyl or an aryl groups that can be removed by mild alkaline hydrolysis, or A may be a β-trichloroethyl group that can be removed by treatment with zink in acetic acid or A may be a β-iodoethyl, a 2-p-tolylsulphonylethyl or a mono- or dihalogenobenzyl group which can be removed by treatment with basic agents, e.g. sodium thiophenolate.

I. A natural of biosynthetic penicillin of the formula



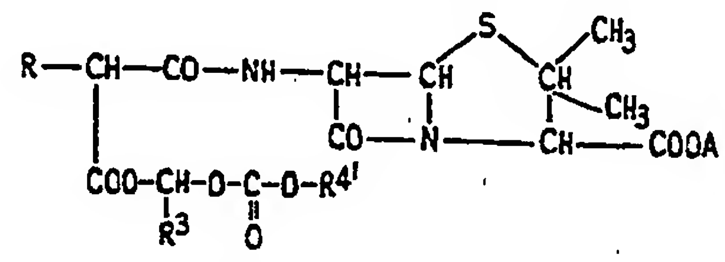
XXI

where R°CO represents the acyl group in the side chain of the natural or biosynthetic penicillin and A has the definition given above is reacted with a phosphorus halide in an inert solvent and suitably in presence of a tertiary amine to give an imino halide compound, which is reacted with a lower alcohol to give an iminoether derivative, which imino ether thereafter is reacted with a compound of the formula

$$\begin{array}{l} R{-}CH{-}CO{-}Z \\ \quad | \\ \quad COO\overset{\displaystyle R^3}{\overset{|}{C}H}{-}O{-}\overset{\displaystyle O}{\overset{\|}{C}}{-}O{-}R^{4'} \end{array}$$

XVI

wherein R, R^3, R^4, $R^{4'}$, and Z have the meanings specified above, and the reaction product treated with water or an alcohol to give a compound of the formula

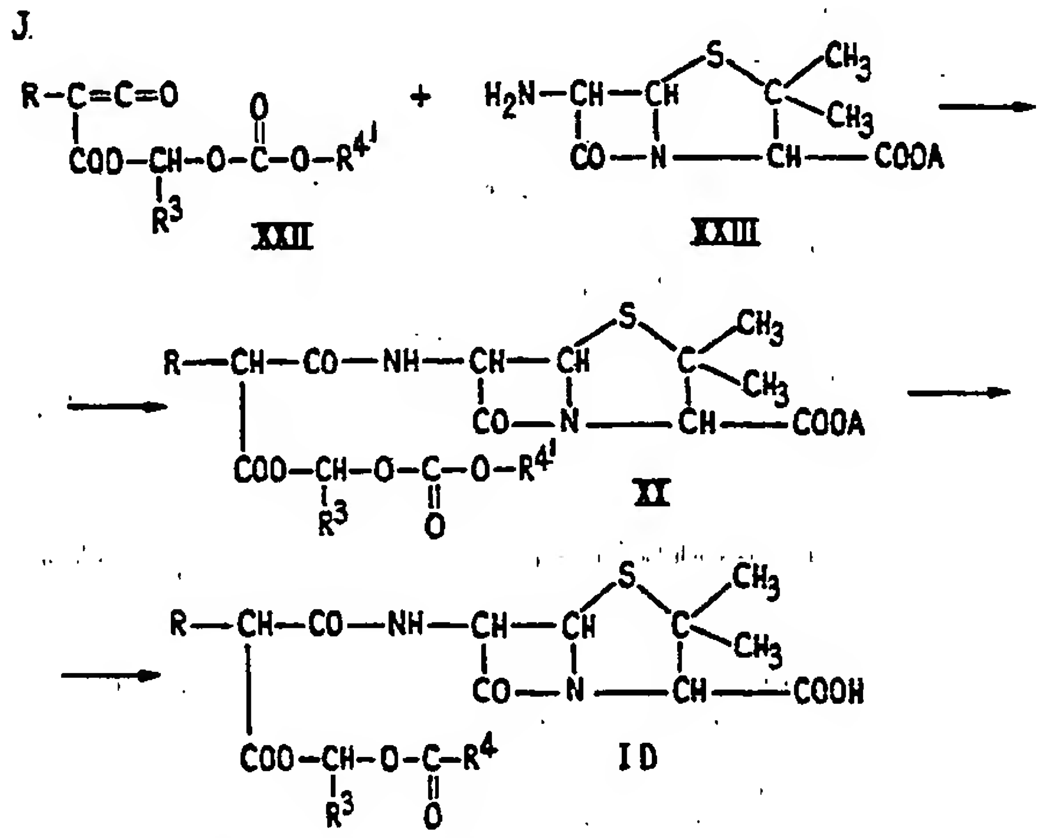


XX

wherein R, R^3, R^4 and A are as defined above which compound is then converted into a compound of the formula I D as is described under H above. In this method the intermediate imino ether compound is directly acylated without isolation of any intermediate products.

The group $R^\circ CO$— in the compound of the formula XXI is an organic acyl group contained in known natural or biosynthetic penicillins. Thus the group R° may be an alkyl, aralkyl or a methyl group substituted with a heterocyclic group and derivatives thereof. Examples of suitable groups R° are heptyl, phenoxymethyl, 2-thienylmethyl, 2-furylmethyl, and benzyl. Examples of suitable phosphorus halides are phosphorus pentachloride, phosphorus pentabromide, phosphorus oxychloride, and phosphorus trichloride. Phosphorus pentachloride is preferred. Examples of suitable alcohols with which the imino halide may be treated are lower alkyl alcohols such as methanol, ethanol, and n-propanol.

J.

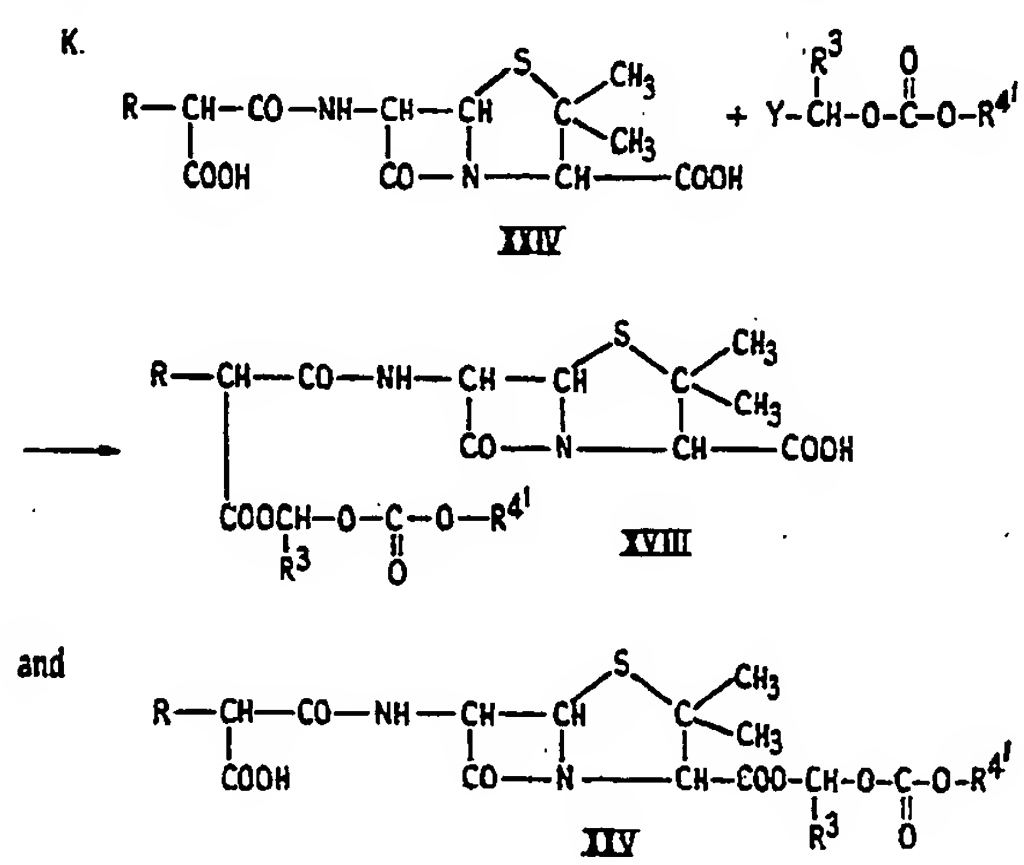


wherein A, R^3, R^4 and $R^{4'}$ are as defined above.

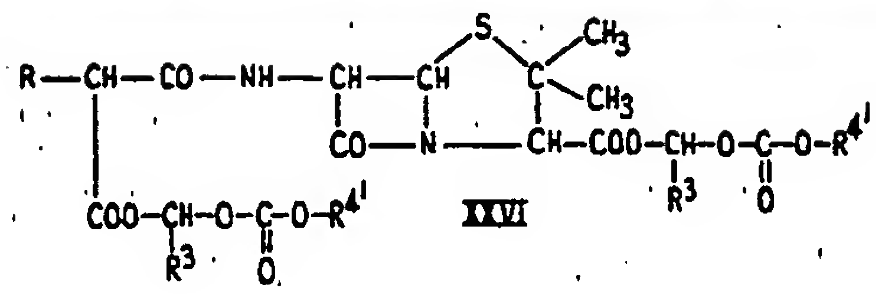
Instead of the activated carboxylic acid derivative XVI of the method G, a ketene derivative of the formula XXII may be used in the acylation reaction as described in the Belgian patent specification 726 421.

Preparation of esters of the penicillins (additional method)

K.



and



XXVI

Treatment of carboxypenicillin with the general formula XXIV, where R has the same meaning as above, with a compound

$$Y-\underset{R^{3}}{\underset{|}{CH}}-O-\underset{O}{\underset{\|}{C}}-C-R^{4'}$$

wherein R^3, $R^{4'}$, and Y have the meanings given above, gives a mixture of the two monoesters XVIII and XXV and the diester XXVI. In the cases where $R^{4'}=R^4$ all these esters are compounds of the invention within the general structure I and they may be used in form of their mixture. When $R^{4'}$ contains groups that are prot·cted, the protecting groups are removed in *per se* known manner in at least one additional step.

If desired the pure compounds XVIII, XXV and XXVI may however, be separated from the mixture by known methods, such as extraction, fractional precipitation or crystallization. The preferred way to prepare the diester XXVI is to treat the carboxypenicillin XXIV with at least two equivalents of

$$Y-\overset{R^{3}}{\overset{|}{CH}}-O-\overset{O}{\overset{\|}{C}}-O-R^{4'}.$$

In the reaction between the carboxypenicillin and

$$Y-\overset{R^{3}}{\overset{|}{CH}}-O-\overset{O}{\overset{\|}{C}}-O-R^{4'}$$

the former is used in form of its salt with inorganic and tertiary organic bases, e.g. as the sodium, potassium, calcium or triethylamine salt, and the reaction is performed in organic solvents such as dimethylformamide, dimethylsulphoxide, acetone, tetrahydrofuran, hexamethylphosphoramide or in mixtures of organic solvents and water, e.g. aqueous acetone or dioxane.

As described above the starting material may be in the form of a salt, for instance a sodium, potassium, calcium or trialkylammonium salt, in some of the methods for the preparation of the compounds of the invention.

In addition, tetraalkylammonium salts and other analogous salts may be used such as salts where the cation has the formula

$$A^1A^2A^3A^4N^{\oplus}$$

in which formula A^1 is a straight or branched alkyl group containing from 3 to 6 carbon atoms, or a substituted or unsubstituted aryl, or substituted or unsubstituted aralkyl group and A^2, A^3 and A^4, which are the same or different, each is a straight or branched alkyl group containing from 1 to 6 carbon atoms, provided that A^2, A^3 and A^4 are alkyl with 3—6 carbon atoms when A^1 is alkyl.

Illustrative examples of suitable combinations of A^1, A^2, A^3 and A^4 in the quaternary ammonium ion $A^1A^2A^3A^4N^{\oplus}$ are given below:

TABLE I

Examples of suitable combinations of the radicals

$A^1 - A^4$ in the $A^1A^2A^3A^4N^{(+)}$ ion

A^1	A^2	A^3	A^4
n-propyl	n-propyl	n-propyl	n-propyl
i-propyl	i-propyl	i-propyl	i-propyl
n-butyl	n-butyl	n-butyl	n-butyl
i-butyl	i-butyl	i-butyl	i-butyl
n-pentyl	n-pentyl	n-pentyl	n-pentyl
n-hexyl	n-hexyl	n-hexyl	n-hexyl
phenyl	methyl	methyl	methyl
phenyl	ethyl	ethyl	ethyl
p-tolyl	ethyl	ethyl	ethyl
p-chlorophenyl	ethyl	ethyl	ethyl

When the radicals A^1—A^4 are all different the resulting ion contains an asymmetric centre and may occur in two enantiomeric forms. Epimeric forms can occur if A^1, A^2, A^3 and/or A^4 contain one or more asymmetric carbon atoms.

Examples of quaternary ammonium ions containing an asymmetric centre are given in Table III below:

TABLE II

Examples of quaternary ammonium ion $A^1A^2A^3A^4N^{(+)}$ containing an asymmetric centre

A^1	A^2	A^3	A^4
benzyl	n-propyl	i-propyl	n-butyl
benzyl	n-propyl	i-propyl	sec.butyl
benzyl	n-propyl	n-butyl	sec.butyl
n-propyl	n-propyl	n-butyl	sec.butyl
n-propyl	n-propyl	n-propyl	sec.butyl
n-propyl	n-propyl	n-propyl	sec.pentyl
n-propyl	n-propyl	n-propyl	sec.hexyl
n-propyl	n-propyl	n-butyl	sec.hexyl

The use as described above of a quaternary salt form of the starting material for the preparation of the compounds of this invention is not previously described in the literature pertaining to this technical field. In this method the preferred cation is the tetraalkylammonium ion, particularly the tetrabutylammonium ion. The preferred solvents are chloroform, methylenechloride and acetone.

The quaternary ammonium salt form of the above described starting material may be prepared by reacting the starting material in question with a quaternary ammonium salt of the formula

$$A^1A^2A^3A^4N^{\oplus}B^{\ominus}$$

wherein A^1, A^2, A^3 and A^4 have the meanings specified above and B is a suitable anion such as $HSO_4^{\ominus}$.$Cl^{\ominus}$ or $CH_3COO^{\ominus}$ to the formation of a quaternary salt of the starting material.

The salts of the formula above which contains B as the anion may be prepared in known manner analogous as described in for instance Belgian patent 751 791. The anion $B^{\ominus}$ is in the preferred embodiment $HSO_4^{\ominus}$.

The following Examples are given to illustrate the invention.

Example 1.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl) monoester sodium salt

a) *By method A*

1) Chloromethylethylcarbonate was prepared by reacting chloromethyl chloroformate (38.7 g, 0.30 mole) with ethanol (13.8 g, 0.30 mole) in dry ether (500 ml) in the presence of pyridine (23.7 g, 0.30 mole). Stirring was continued at room temperature for 3 hours. After filtration and evaporation the residue was distilled to give a colourless liquid (33.0 g, 79%. Bp_{14}: 48—50°C).

2. To a stirred and ice-cooled suspension of potassium phenylacetate (43.5 g, 0.25 mole) in dry dimethyl sulphoxide (80 ml) was added dropwise chloromethylethylcarbonate (27.7 g, 0.20 mole). Stirring was continued at room temperature for 18 hours. The mixture was poured into an ice-cooled 0.5 N sodium bicarbonate solution (500 ml) and after stirring for 20 minutes the mixture was extracted with ethyl acetate (3x150 ml). The combined organic phase was washed with cold water, dried over anhydrous magnesium sulphate, and evaporated. The crude oil (44.9 g, 94%) was used in the next step.

3) To a stirred solution of N-isopropylcyclohexylamine (8.4 g, 60 mmole) in dry tetrahydrofuran (60 ml) was added (N_2 atmosphere, −78°C) a 1.5 N solution of n-butyllithium in hexane (40 ml, 60 mmole). After 15 minutes, a solution of the above obtained ethoxycarbonyloxymethyl phenylacetate (13.0 g, 54.5 mmole) in dry tetrahydrofurane (40 ml) was added dropwise during one hour, and then an excess of powdered dry ice was added and stirring was continued for 15 minutes. The solution was added dropwise to ice-cooled 2N hydrochloric acid (100 ml) and, after stirring 15 minutes, this mixture was extracted with chloroform (3x75 ml). The combined organic phase was washed with cold water, water (100 ml) was added and pH was adjusted to 7.5 with 1N sodium bicarbonate solution. The organic phase was washed with water and, after washing with chloroform, the combined water phase was added to diethyl ether and pH was adjusted to 1.0 with 2N hydrochloric acid. The water phase was washed with ether, and the combined organic phase was washed with water and dried. Evaporation gave a crystalline residue (9.7 g, 63%) which was identified as phenylmalonic acid ethoxycarbonyloxymethyl monoester.

The infrared (IR) spectrum (KBr disc) had absorption maximum (cm^{-1}) at 3700—2150 (carboxyl OH): 1755 (ester and carbonate C=O); 1690 (carboxyl C=O). The nuclear magnetic resonance spectrum (NMR) in deuterochloroform showed absorptions (p.p.m. (δ) from tetramethylsilane) at 9.50 (s, COOH); 7.33 (s, C_6H_5); 5.79 (s, OCH_2O); 4.70 (s, $C_6H_5\mathit{CH}CO$); 4.19 (q, $O\mathit{CH_2}CH_3$); 1.23 (t, $OCH_2\mathit{CH_3}$).

4) The phenylmalonic acid ethoxycarbonyloxymethyl-monoester (1.13 g, 4.0 mmole) was stirred with thionyl chloride (1.67 g, 14 mmole) at 65°C for one hour and then the reaction mixture was evaporated to dryness with dry benzene (25 ml) four times.

The crude acid chloride (1.20 g, 4.0 mmole) was dissolved in dry methylene chloride (5 ml) and added dropwise to a stirred and ice-cooled solution of 6-amino-penicillanic acid benzylhydrylester p-toluenesulphonate (2.22 g, 4.0 mmole) and dry triethylamine (1.01 g, 10.0 mmole) in dry methylene chloride (35 ml). Stirring

was continued for 90 minutes at 0°C, then cold water (40 ml) was added and pH was adjusted to 2.0 with 2N hydrochloric acid. The organic phase was separated and washed successively with saturated sodium bicarbonate solution and sodium chloride solution. After drying and evaporating the residue (2.4 g) was chromatographed on a silica gel column (40 g) prepared in dry benzene. The residue was applied dissolved in a minimum amount of benzene, and eluted with gradient technique, isopropylether-acetone (8:2) was used as the second solvent. The fractions collected were checked by thin layer chromatography (TLC) on silica gel plates using the same solvent mixture. In this way a white foam (1.30 g, 50%) was isolated from one of the middle fractions of the eluate. It showed only one spot on TLC.

IR(KBr): 1780—1740 (β-lactam, ester and carbonate C=O); 1680 (amide C=O). NMR($CDCL_3$): 7.38 (s, 3 C_6H_5); 6.94 (s, $CH(C_6H_5)_2$); 5.79 (s, OCH_2O); 5.80—5.40 (m, 5-H and 6-H); 4.66 (d, C_6H_5CHCO); 4.51 (d, 3-H); 4.20 (q, OCH_2CH_3); 1.60—1.10 (m, OCH_2CH_3, and gem. CH_3).
Analysis: Calculated for $C_{34}H_{34}O_9N_2S$ (646.73): C 63.14; H 5.30; O 22.27; N 4.33; S 4.96. Found: C 63.28, H 4.32, O 22.17; N 4.18; S 4.86.

5) The above obtained 6-(α-carboxyphenylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl)-3-(benzhydryl) diester (1.15 g, 1.8 mmole) was dissolved in a 1:1 mixture of ethyl acetate and ethanol (10 ml) and added to a prehydrogenated palladium-charcoal catalyst (1.0 g, Pd cont. 10%) in a mixture of ethanol (5 ml) and water (5 ml) containing sodium bicarbonate (0.15 g, 1.8 mmole). Hydrogenation was continued at normal pressure and room temperature for 2 hours, then the catalyst was filtered off, ethanol and ethyl acetate was removed at reduced pressure and the resulting mixture was washed with ethyl acetate. Ethyl acetate (10 ml) was added and pH was adjusted to 2.0 with 2N hydrochloric acid. The organic phase was dried, and a 2N solution of sodium 2-ethylhexanoate (1.0 ml, 2 mmole) was added, the solution was evaporated to a minimum volume, and the sodium salt was precipitated with dry ether. The filtered product (0.50 g, 55%) showed only one spot on TLC in butanone-pyridine-water-acetic acid (70:15:15:2) system and was identical with, but purer than, the substance prepared by method E.

IR(KBr): 1780—1740 (β-lactam, ester and carbonate C=O); 1675 (amide C=O); 1610 (carboxyl C=O). NMR(D_2O): 7.40 (s, C_6H_5); 5.80—5.60 (m, 5-H, 6-H and OCH_2O); 4.30 (d, 3-H); 4.10 (q,OCH_2CH_3); 1.51 (d, gem, CH_3); 1.12 (t, OCH_2CH_3).
Analysis: Calculated for $C_{21}H_{23}O_9N_2SNa$ (502.49): N 5.58; S 6.38; Na 4.58. Found: N 5.45; S 6.22; Na 4.68.

The degree of hydrolysis of all 6-(α-carboxyphenylacetamido) penicillanic acid derivatives described herein was studied in Sörensen's buffer solution (B), in 25% human serum (H) and in 5% rat serum (R) in the presence of 10% dimethyl sulphoxide, the pH of each mixture being adjusted to 6.8. The mixtures were incubated at 37°C, samples being taken at different intervals after three (B3 and H3) and two (R2) hours respectively, aliquots spotted onto paper tapes and the reaction mixture components were separated chromatographically, using butanol-ethanol-water (4:1:1) solvent system. The concentration of the liberated free 6-(α-carboxyphenylacetamido) penicillanate was quantitatively estimated by microbiological detection (Bacillus Subtilis), against simultaneously ran standards. The degree of hydrolysis of the substance described in this example was: B3=24.2%; H3=37.5%; R2=95%.

b) *By method G*

The phenylmalonic acid ethoxycarbonyloxymethylmonoester chloride (3.0 g 10 mmole) in dry ether (5 ml) was added to a well stirred and ice-cooled solution of sodium 6-aminopenicillinate, prepared by suspending 6-aminopenicillanic acid (3.24 g, 15 mmole) in 50% acetone (50 ml) and adjusting the pH to 7.0 with 2N sodium hydroxide. During the addition of acid chloride, the pH was kept constant at 7.0 by addition of alkali. Stirring was continued for one hour at 0°C, then the organic solvents were distilled off at reduced pressure and the remaining water phase was washed with ether. The pH was adjusted to 4.5 with 2N hydrochloric acid, the precipitate was filtered off and the filtrate was acidified to pH 2.2 in the presence of ether (50 ml). The organic phase was washed with water, and then extracted by addition of water (50 ml) and adjusting the pH to 7.0 with 2N sodium hydroxide solution. The ether free water phase was freeze-dried to give a colourless powder (3.1 g, 62%) showing a main spot on TLC (Butanone-pyridine-water-acetic

acid system) besides a minor quantity of 6-(α-carboxyphenylacetamido)-disodium penicillinate. This substance was by spectral, analytical and hydrolysis data identical with the substance prepared by method A.

Example II.

Preparation of 6-(α-carboxy-3-thienylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl) monoester sodium salt

1) Chloromethylethylcarbonate (6.9 g, 50 mmole) was added dropwise to an ice-cooled suspension of potassium 3-thienylacetate (10.8 g, 60 mmole) in dry dimethyl sulfoxide (20 ml) and the reaction mixture was stirred at room temperature for 20 hours, followed by working up in a similar way as in example Ia. The residual oil (11.1 g, 91%) was uniform according to TLC analysis in isopropylether-acetone (8:2) system.

2) A solution of the above obtained ethoxycarboxyloxymethyl-(3-thienyl)-acetate (7.1 g, 29 mmole) in dry tetrahydrofuran (20 ml) was added dropwise (−78°C, N_2 atmosphere) during one hour to a solution of lithium N-isopropyl-cyclohexylamide, prepared as in example Ia by reacting N-isopropylcyclohexylamine (4.2 g, 30 mmole) with n-butyllithium (30 mmole). Powdered dry ice was added, and after 10 minutes the reaction mixture was worked up as in example Ia to give a product (6.1 g, 73%), identified as 3-thienylmalonic acid ethoxycarbonyloxymethyl monoester. IR(KBr): 3700—2100 (carbonyl OH); 760 (ester and carbonate C=0); 1690 (carboxyl C=0). NMR($CDCl_3$): 9.51 (s, COOH); 7.35—7.05 (m, C_4H_3S); 5.80 (s, OCH_2O); 4.83 (s, C_4H_3SCHCO); 4.19 (q, OCH_2CH_3); 1.23 (t, OCH_2CH_3).

3) 3-Thienylmalonic acid ethoxycarbonyloxymethyl monoester chloride was prepared as in example Ia from the above obtained acid (1.73 g, 6.0 mmole) and thionyl chloride (1.67 g, 14 mmole). After working up in the usual manner (codistillation with dry benzene) the crude oil (1.84 g, 6.0 mmole) was dissolved in ether (5 ml) and added dropwise to an ice-cooled 50% acetone solution of sodium 6-aminopenicillinate, prepared from 6-aminopenicillanic acid (1.95 g, 9 mmole) according to the description given in example Ib. Working up and freeze-drying as in example Ib gave a powder (1.8 g, 59%) which showed a main spot in TLC (butanone-pyridine-water-acetic acid system) besides a minor quantity of 6-(α-carboxy-3-thienylacetamido) disodium penicillinate. IR(KBr): 1780—1740 (β-lactam, ester and carbonate C=0); 1680—1670 (amide C=0); 1610 (carboxyl C=0). NMR(D_2O): 7.35—7.05 (m, C_4H_3S); 5.80—5.60 (m, 5-H, 6-H and OCH_2O); 4.30 (d, 3-H); 4.11 (q, OCH_2CH_3); 1.50 (d, gem. CH_3); 1.12 (t, OCH_2CH_3).

Analysis: Calculated for $C_{19}H_{21}O_9N_2S_2Na$ (508.52): N 5.51; S 12.61; Na 4.52. Found: N 5.38; S 12.52; Na 4.62.

The degree of hydrolysis of this compound was: B3=11.5%; H3=32.8%; R2=104%.

Example III.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(phenoxycarbonyloxymethyl) monoester sodium salt

1) Chloromethylchloroformate (38.7 g, 0.30 mole) in dry ether (150 ml) was added dropwise to a stirred and ice-cooled solution of phenol (28.2 g, 9.30 mole) and pyridine (23.7 g, 0.30 mole) in dry ether (400 ml). Stirring was continued for 16 hours, then the pyridine hydrochloride was filtered off the filtrate was evaporated and the residue (45.3 g) was distilled to give chloromethylphenylcarbonate as a colourless liquid (40.7 g, 73%. $Bp_{0.04}$: 65—68°C).

2) Phenylacetic acid phenoxycarbonyloxymethyl ester was prepared in a similar way as in example Ia from chloromethylphenylcarbonate and potassium phenylacetate, and the crude ester was treated −78°C under nitrogen with lithium N-isopropylcyclohexylamide followed by the addition of dry ice, as described in example Ia. Working up in the usual manner gave phenylmalonic acid phenoxycarbonyloxymethyl monoester, which was converted to its acid chloride by heating with thionyl chloride, excess reagent being removed by codistillation with dry benzene.

3) A solution of phenylmalonic acid phenoxycarbonyloxymethyl monoester chloride (1.74 g, 5.0 mmole) in dry methylene chloride (5 ml) was added to a stirred and ice-cooled solution of triethylamine (1.21 g, 12.5 mmole) and 6-aminopenicillanic acid benzhydryl ester p-toluene sulphonate (2.77 g, 5.0 mmole) in dry methylene chloride (45 ml). After stirring for 90 minutes at 0°C, the reaction mixture was worked up as in example Ia to give a yellowish foam (3.34 g) which was chromatographed on a silica gel column (50 g), prepared in dry benzene.

Elution with gradient technique, using isopropylether-acetone (8:2) as the second solvent, gave a main fraction containing 6-(α-carboxyphenylacetamido)-penicillanic acid α-(phenoxycarbonyloxymethyl)-3-benzhydryldiester (1.84 g, 53%) isolated as a white foam, which was pure according to TLC analysis. IR(KBr): 1780—1740 (β-lactam, ester and carbonate C=0); 1675 (amide C=0). NMR($CDCl_3$): 7.45—7.10 (m, 4 C_6H_5); 6.95 (s, $(C_6H_5)_2CH$); 5.80 (s, OCH_2O); 5.80—5.40 (m, 5-H and 6-H); 4.67 (d, C_6H_5CHCO); 4.51 (d, 3-H); 1.40 (s, gem CH_3).

Analysis: Calculated for $C_{38}H_{34}O_9N_2S$ (694.78): C 65.69; H 4.93; O 20.73; N 4.03; S 4.62. Found: C 65.75; H 4.96; O 20.62; N 4.00; S 4.58.

4) The diester (1.60 g, 2.30 mmole) was hyrogenated over palladiumcharcoal as in example Ia, and the sodium salt (0.82 g, 65%) was precipitated in the usual manner with sodium 2-ethylhexanoate. The substance showed only one spot on TLC. IR(KBr): 1780—1740 (β-lactam, ester and amide C=0); 1680 (amide C=0) 1610 (carboxyl C=0). NMR(D_2O): 7.45—7.10 (m, 2 C_6H_5); 5.80—5.60 (m, 5-H, 6-H and OCH_2O); 4.30 (s, 3-H); 1.50 (d, gem, CH_3).

Analysis: Calculated for $C_{25}H_{23}O_9N_2SNa$ (550.52): N 5.9; S 5.83; Na 4.18. Found: N 4.96; S 5.62; Na 4.48.

Degree of hydrolysis: B3=29.2%; H3=42.5%; R2=117%.

Example IV

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid 3-(1′-ethoxycarbonyloxyethyl) monoester sodium salt

a) *By method A, indirect route*

1) To a solution of 6-aminopenicillanic acid 3-(1′-ethoxycarbonyloxyethyl) ester p-toluenesulphonate (3.24 g, 8.0 mmole) in dry methylene chloride (70 ml) containing triethylamine (2.01 g, 20.0 mmole) was added dropwise, with stirring and ice-cooling, phenylmalonic acid monobenzylester chloride (2.31 g, 8.0 mmole) in dry methylene chloride (10 ml). Stirring was continued for 90 minutes at 0°C and then the mixture was worked up and chromatographed on silica gel (100 g) according to the description given in example Ia. The diester (3.03 g, 65%) was isolated as a foam, which was pure according to TLC analysis. IR($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0): 1680 (amide C=0). NMR($CDCl_3$): 7.28 (d, 2 C_6H_5); 6.77 (q, $OCH(CH_3)O$); 5.80—5.40 (m, 5-H and 6-H); 5.16 (s, $C_6H_5CH_2O$); 4.61 (d, C_6H_5CHCO); 4.42 (d, 3-H); 4.20 (q, OCH_2CH_3); 1.60—1.10 (m, gem, CH_3, OCH_2CH_3 and $OCH(CH_3)O$).

Analysis: Calculated for $C_{29}H_{32}O_9N_2S$ (584.66): C 59.58; H 5.52; O 24.63; N 4.79; S 5.49. Found: C 59.66; H 5.60; O 24.34; N 4.64; S 5.32. Degree of hydrolysis: B3 = < 1%; H3 = < 1%; R2 = 47.5%.

2) The above obtained diester (2.92 g, 5.0 mmole) was dissolved in ethanol (10 ml) and added to a prehydrogenated palladium-charcoal catalyst (3.0 g, Pd cont. 10%) in a mixture of ethanol sodium bicarbonate (0.42 g, 5.0 mmole). Hydrogenation was continued at normal pressure and room temperature for 2 hours, then the catalyst was filtered off and ethanol was removed at reduced pressure. Ether (25 ml) was added, pH was adjusted to 2.2 with 2N hydrochloric acid and the organic phase was separated and washed with water. Water (25 ml) was added, pH was adjusted to 7.0 with 2N sodium hydroxide solution and the ether-free water phase was freeze-dried to give a white powder (1.60 g, 62%), which was pure according to TLC analysis (butanone-pyridine-water- acetic acid system). IR(KBr): 1780—1760 (β-lactam, ester and carbonate C=0); 1675 (amide C=0); 1615 (carboxyl C=0). NMR (D_2O): 7.35 (s, C_6H_5); 6.71 (q, $OCH(CH_3)O$); 5.70—5.60 (m, 5-H and 6-H); 4.4? (s, 3-H); 4.15 (q, OCH_2CH_3); 1.50—1.10 (m, gem, CH_3, OCH_2CH_3 and $OCH(CH_3)O$).

Analysis: Calculated for $C_{22}H_{25}O_9N_2SNa$ (516,51): N 5.42; S 6.21; Na 4.45. Found: N 5.36; S 6.16; Na 4.74. Degree of hydrolysis: B3 = 3.5%; H3 = 13.5%; R2 = 83.5%.

b) *By method A, direct route*

Crude 6-aminopenicillanic acid 3-(1′-ethoxycarbonyloxyethyl) ester, prepared according to example IV from 6-phenylacetamidopenicillanic acid 3-(1′-ethoxycarbonyloxyethyl) ester (2.70 g, 6.0 mmole), was dissolved in 50% acetone (20 ml) and acylated with phenylmalonic acid monochloride (0.80 g, 4.0 mmole) using the same reaction conditions and working up procedure as in example Ib. Freeze-drying of the ether-free water phase gave the title compound as powder (0.74 g, 30%). This substance was by spectral, analytical and hydrolysis data identical with the substance prepared by the indirect route (IVa), but had a lower purity.

c) *By method A, ketene route*

A dry methylene chloride (5 ml) solution of phenyl (chlorocarbonyl) ketene (0.45 g, 2.5 mmole), prepared from phenylmalonic acid and phosphorus pentachloride (C.A. 73, 25451 t, 1970), was added dropwise to a stirred solution of 6-aminopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester p-toluenesulphonate (1.01 g, 2.0 mmole) in dry methylene chloride (10 ml) containing triethylamine (0.22 g, 2.2 mmole) at −20°C. Stirring was continued for 90 minutes at 0°C, then water (20 ml) was added and the pH was adjusted to 2.2 with 2N hydrochloric acid. The methylene chloride solution was then extracted with water (15 ml), the pH being adjusted to 7.0 with 2N sodium hydroxide solution. The water phase was freeze-dried to give the title compound as a powder (0.66 g, 64%), which was by spectral, analysis and hydrolysis data identical with the substance prepared by the indirect route (IVa), but had a lower purity.

d) *By method C*

To a stirred and ice-cooled solution of tetrabutylammonium hydrogen sulphate (17.0 g, 50 mmole) in water (25 ml) was added chloroform (50 ml) and the pH was adjusted to 7.0 with 2N sodium hydroxide solution. 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzylmonoester potassium salt (25.3 g, 50 mmole) was added portionwise, then the organic phase was separated and, after drying with anhydrous magnesium sulphate, α-chlorodiethylcarbonate (7.6 g, 50 mmole) was added. The solution was left at 40°C for 16 hours.

The reaction mixture was evaporated, water (150 ml) was added and the product was extracted with ether (3×100 ml). The collected organic phase was washed successively with water, saturated sodium bicarbonate solution and water. Drying and evaporation gave an oily residue (17.4 g) which was chromatographed using silica gel (250 g) and the usual technique. The diester (8.9 g, 30.5%) was isolated as a foam and it was in all respects identical with the diester prepared by the indirect route (IVa).

2) Hydrogenation of this substance as previously described in this example gave the title compound (5.4 g, 69%), identical by spectral, analytical and hydrolysis data with the substance under VIa.

Example V.

Preparation of 6-(α-carboxy-3-thienylacetamido)-penicillanic acid 3-(1'-ethoxycarbonyloxyethyl) monoester sodium salt

3-Thienylacetic acid (7.1 g, 50 mmole) was stirred with thionyl chloride (12.0 g, 100 mmole) at 40°C for one hour and then codistilled with dry benzene (4×50 ml). The crude acid chloride (50 mmole) was added dropwise to an ice-cooled solution of benzyl alcohol (5.4 g, 50 mmole) in pyridine (30 ml) and the mixture was stirred for 18 hours.

The reaction mixture was poured into 50 ml ice-water and after acidifying with hydrochloric acid, it was extracted with chloroform. The extract was washed with saturated sodium bicarbonate and water, dried and evaporated. The residue was distilled to give pure benzyl (3-thienyl) acetate (6.75 g, 58%, $Bp_{0.1}$, 143—144°C).

This ester (4.65 g, 20 mmole) was converted to 3-thienylmalonic acid monobenzylester (4.20 g, 76%) using lithium N-isopropylcyclohexylamide and carbon dioxide in a similar way as in example Ia.

Its acid chloride (1.92 g, 6.5 mmole), was used to acylate 6-aminopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester p-toluenesulphonate (3.28 g, 6.5 mmole) using the same method as in example VIa. Chromatography on silica gel (60 g) gave pure 6-(α-carboxy-3-thienylacetamido)-penicillanic acid α-benzyl-3-(1'-ethoxycarbonyloxyethyl) diester (2.23 g, 58%). IR($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680—1670 (amide c=0). NMR($CDCl_3$); 7.45—7.05 (m, C_6H_5 and C_4H_3S); 6.77 (q, OCH(CH_3)O); 5.80—5.40 (m, 5-H and 6-H); 5.16 (s, $C_6H_5CH_2O$); 4.77 (d, C_4H_3SCHCO); 4.42 (d, 3-H); 4.20 (q, OCH_2CH_3); 1.60—1.10 (m, gem. CH_3, OCH_2CH_3 and OCH(CH_3)O).

Analysis: Calculated for $C_{27}H_{30}O_9N_2S_2$ (590.69): C 54.90; H 5.12; O 24.38; N 4.74; S 10.86. Found C 54.85; H 5.22; O 24.54; N 4.62; S 10.82. Degree of hydrolysis: B3 = < 1%; H3 = < 1%; R2 = 39.8%.

4) The diester (2.09 g, 3.5 mmole) was hydrogenated over palladiumcharcoal (2.0 g) as described in example IVa. The freeze-dried product (1.17 g, 64%) showed only one spot on TLC (butanone-pyridine-water-acetic acid system). IR: (KBr) 1780—1760 (β-lactam, ester and carbonate C=0); 1675 (amide C=0); 1610

(carboxyl C=0) NMR (D_2O): 7.35—7.05 (m, C_4H_3S); 6.71 (q, $OCH(CH_3)O$); 5.70—5.60 (m, 5-H and 6-H); 4.43 (s, 3-H); 4.15 (q, OCH_2CH_3); 1.50—1.10 (m, gem, CH_3, OCH_2CH_3 and $OCH(CH_3)O$). Analysis: Calculated for $C_{20}H_{23}O_9N_2S_2Na$ (522.53): N 5.36; S 12.27; Na 4.40. Found: N 5.22; S 12.08; Na 4.62. Degree of hydrolysis: B3 = 4.9%; H3 = 15.3%; R2 = 92.6%.

Example VI.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl)-3-(1'-ethoxycarbonyloxyethyl) diester

a) *By method A, direct acylation route*

Crude 6-aminopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester, prepared as in example IVb from 6-phenylacetamidopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester (4.05 g, 9.0 mmole), was acylated in methylene chloride (70 ml) solution, as described in example Ia, with phenylmalonic acid ethoxycarbonyloxymethyl monoester chloride (2.70 g, 9.0 mmole). After working up, the crude oil was chromatographed on silica gel (100 g) to give the title compound, isolated as a foam (0.75 g, 14%) from one of the middle fractions of the eluate. IR ($CHCL_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1685—1675 (amide C=0); NMR ($CDCl_3$): 7.40 (s, C_6H_5); 6.80 (q, $OCH(CH_3)O$); 5.81 (s, OCH_2O); 5.80—5.40 (m, 5-H and 6-H); 4.61 (d, C_6H_5CHCO); 4.41 (d, 3-H); 4.23 (q, OCH_2CH_3); 1.60—1.10 (m, gem, CH_3, OCH_2CH_3 and $OCH(CH_3)O$).
Analysis: Calculated for $C_{26}H_{32}O_{12}N_2S$ (596.67). C 52.34; H 5.41; O 32.18; N 4.70; S 5.38. Found: C 52.44; H 5.48; O 31.88; N 4.62; S 5.44. Degree of hydrolysis: B3 = 3.8%; H3 = 12.7%; R2 = 46.5%.

b) *By method A, addition route*

Prosphorus pentachloride (2.07 g, 9.9 mmole) was added during 5 min. with vigorous stirring to a solution of 6-phenylacetamidopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester (4.05 g, 9.0 mmole) and N,N-dimethylaniline (3.7 ml) in dry methylene chloride (40 ml) at −25°C under dry nitrogen. After 1.5 hours dry methanol (36 ml) was added while keeping the temperature at −25° — −30°C. After an additional 2.5 hours N,N-dimethylaniline (5.4 ml) was added, followed by the dropwise addition during one hour of phenylmalonic acid ethoxycarbonyloxymethyl monoester chloride (3.25 g, 10.8 mmole) in methylene chloride (10 ml). The mixture was then stirred for 16 hours at −20°C. Water (27 ml) was added, pH was adjusted to 2.0 with 2N hydrochloric acid and the organic phase was separated and washed successively with saturated sodium bicarbonate solution and sodium chloride solution. After drying and evaporating, the residue (8.70 g) was chromatographed on silica gel (100 g). The title compound was isolated as a foam (2.30 g, 43%) from one of the middle fractions of the eluate. It showed the same spectral, analytical and hydrolysis data as the sample prepared under a).

Example VII.

Preparation of 6-(α-carboxy-3-thienylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl)-3-(1'-ethoxycarbonyloxyethyl) diester

3-Thienylmalonic acid ethoxycarbonyloxymethyl monoester chloride (0.92 g, 3.0 mmole) was reacted with 6-aminopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester p-toluenesulphonate (1.51 g, 3.0 mmole) in methylene chloride solution in the presence of a triethylamine (0.76 g, 7.5 mmole) in a similar way as described in example Ia. After working up and chromatography on silica gel (40 g) the title compound was isolated as a foam (0.99 g, 55%) from one of the middle fractions of the eluate. IR($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR($CDCl_3$): 7.35—7.05 (m, C_4H_3S); 6.78 (q, $OCH(CH_3)O$); 5.81 (s, OCH_2O); 5.80—5.40 (m, 5-H and 6-H); 4.78 (C_4H_3SCHCO); 4.41 (d, 3-H); 4.23 (q, OCH_2CH_3); 1.50—1.10 (m, gem. CH_3, OCH_2CH_3 and $OCH(CH_3)O$).
Analysis: Calculated for $C_{24}H_{30}O_{12}N_2S_2$(602.66). C 47.83; H 5.02; O 31.86; N 4.65; S 10.64. Found; C 47.62; H 4.88; O 31.58; N 4.44; S 10.28. Degree of hydrolysis: B3 = 2.8%; H3 = 13.6%. R2 = 51.5%.

Example VIII.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-phenyl-3-(1'-ethoxycarbonyloxyethyl) diester

6-aminopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester p-toluenesulphonate (1.26 g, 2.5 mmole) was acylated in methylene chloride solution

as previously described with phenylmalonic acid monophenylester chloride (0.69 g, 2.5 mmole). Working up and chromatography on silica gel (30 g) gave the title compound as a foam (0.87 g, 61%), from one of the middle fractions of the eluate. IR($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR($CDCl_3$): 7.40—7.10 (m, 2 C_6H_5); 6.78 (q, O*CH*(CH_3)O); 5.80—5.40 (m, 5-H and 6-H); 4.60 (d, C_6H_5*CH*CO); 4.42 (d, 3-H); 4.22 (q, O*CH*$_2$$CH_3$); 1.60—1.10 (m, gem. CH_3, OCH_2*CH*$_3$, and OCH(*CH*$_3$)O).
Analysis: Calculated for $C_{28}H_{30}O_9N_2S$ (570.64); C 58.94; H 5.30; O 25.23; N 4.91; S 5.62. Found: C 58.72; H 5.16; O 24.92; N 4.84; S 5.55. Degree of hydrolysis: B3 = <1%; H3 = 4.2%; R2 = 58.5%.

Example IX.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(5'-indanyl)-3-(1'-ethoxycarbonyloxyethyl) diester

6-aminopenicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester p-toluenesulphonate (2.02 g, 4.0 mmole) was acylated with phenylmalonic acid 5'-indanylmonoester chloride (1.26 g, 4.0 mmole) using the same method as in the previous example. After chromatography on silica gel (50 g) the title compound was isolated as a foam (1.51 g, 62%) from one of the middle fractions of the eluate. IR($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR($CDCl_3$): 7.40—6.90 (m, C_6H_5 and Indanyl-H); 6.78 (q, O*CH*(CH_3)O); 5.80—5.40 (5-H and 6-H); 4.61 (d, C_6H_5*CH*CO); 4.41 (d, 3-H); 4.20 (q, O*CH*$_2$$CH_3$); 2.86 (t, indanyl-H); 2.30—1.90 (m, indanyl-H); 1.50—1.10 (m, gem. CH_3, OCH_2*CH*$_3$ and OCH(*CH*$_3$)O);
Analysis: Calculated for $C_{31}H_{34}O_9N_2S$ (610.70): C 60.97; H 5.61; O 23.58; N 4.59; S 5.25. Found: C 60.52; H 5.38; O 23.72; N 4.62; S 5.18. Degree of hydrolysis: B3 = <1%; H3 = 12.3%; R2 = 65.8%.

Example X.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α, 3-(ethoxycarbonyloxymethyl) diester

To a stirred and ice-cooled suspension of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl) monoester sodium salt (1.51 g, 3.0 mmole) in dry dimethyl formamide (6.0 ml) containing potassium iodide (0.01 g) was added dropwise chloromethylethylcarbonate (0.49 g, 3.5 mmole).

After stirring for 16 hours at room temperature, the reaction mixture was poured into a saturated sodium bicarbonate solution (10 ml) and after stirring for 10 minutes the mixture was extracted with ether (3x10 ml). The collected organic phase was washed with water, dried and evaporated to give an oily residue (1.40 g) which was chromatographed on silica gel (40 g) in the usual way. The title compound was isolated as a foam (0.26 g, 15%) from one of the middle fractions of the eluate. IR($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR($CDCl_3$): 7.39 (s, C_6H_5); 5.80 (s, OCH_2O); 5,80—5.40 (m, 5-H and 6-H); 4.60 (d, C_6H_5*CH*CO); 4.42 (d, 3-H); 4.21 (q, O*CH*$_2$$CH_3$); 1.50—1.10 (m, gem. CH_3, OCH_2*CH*$_3$).
Analysis: Calculated for $C_{25}H_{30}O_{12}N_2S$ (582.60); C 51.54; H 5.19; O 32.95; N 4.81; S 5.55. Found: C 51.46; H 5.08; O 32.72; N 4.72; S 5.36. Degree of hydrolysis: B3 = 13.8%; H3 = 29.2%; R2 = 88.2%.

Example XI.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid 3-(ethoxycarbonyloxymethyl) monoester sodium salt

1) To a stirred and ice-cooled suspension of 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzyl monoester potassium salt (20.2 g, 40 mmole) in dry dimethyl sulphoxide (32.5 ml) was added dropwise chloromethylethylcarbonate (5.5 g, 40 mmole).

After stirring for 16 hours at room temperature, the reaction mixture was poured into an ice-cooled saturated sodium bicarbonate solution (150 ml). After stirring for 10 minutes this mixture was extracted with ethyl acetate (3x75 ml). The organic phase was washed with water, dried and evaporated to give an oil (18.5 g) which was chromatographed on silica gel (250 g) using the usual solvent system. In this way 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzyl-3-(ethoxycarbonyloxymethyl) diester (9.8 g, 43%) was isolated from the main fraction of the eluate. IR ($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR($CDCl_3$): 7.30 (d, 2 C_6H_5); 5.78 (s, OCH_2O); 5.80—5.40 (m, 5-H

and 6-H); 5.16 (s, $C_6H_5CH_2O$); 4.60 (d, C_6H_5CHCO); 4.42 (d, 3-H); 4.22 (q, OCH_2CH_3); 1.60—1.10 (m, gem. CH_3 and OCH_2CH_3). Analysis: Calculated for $C_{28}H_{30}O_9N_2S$ (570.64): C 58.98; H 5.30; O 25.23; N 4.91; S 5.62. Found: C 58.76; H 5.14; O 25.42; N 4.82; S 5.46. Degree of hydrolysis: B3 = < 1%; H3 = < 1%; R2 = 64.5%.

2) The diester (9.7 g, 17 mmole) was hydrogenated over palladium charcoal (10.0 g, Pd cont. 10%) using the method described in example VIa. The freeze-dried product (6.0 g, 70%) was a white powder which showed only one spot on TLC (butanone-pyridine-water-acetic acid system). IR (KBr): 1780—1740 (β-lactam, ester and carbonate C=0); 1685—1675 (amide C=0); 1615—1605 (carboxyl C=0). NMR (D_2O): 7.33 (s, C_6H_5): 5.70—5.60 (m, OCH_2O, 5-H and 6-H); 4.45 (s, 3-H); 4.17 (q, OCH_2CH_3); 1.50—1.10 (m, gem. CH_3 and OCH_2CH_3); Analysis: Calculated for $C_{21}H_{23}O_9N_2SNa$ (502.48): N 5.58; S 6.38; Na 4.58. Found N 5.31; S 6.38; Na 4.88. Degree of hydrolysis: B3 = 18.2%; H3 = 37.5%; R2 = 109%.

Example XII.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid 3-(2′-methylaminoethoxycarbonyloxymethyl) ester salt

1) From chloroformic acid benzylester (34.12 g, 0.20 mole) and 2-methylaminoethanol (37.55 g, 0.50 mole) in dry ether (300 ml) N-benzyloxycarbonylmethyl amino ethanol (40.3 g, 96%) was prepared in the usual way.

2) Chloromethyl chloroformate (10.2 g, 72 mmole) in dry ether (50 ml) was added dropwise to a stirred and ice-cooled solution of N-benzyloxycarbonylmethylaminoethanol (15.0 g, 72 mmole) and dry pyridine (5.7 g, 72 mmole) in dry ether (100 ml). Stirring was continued for 1.5 hours after which the solution was filtered. The filtrate was washed with 1N hydrochloric acid (50 ml), water (50 ml) and 0.5 N sodium bicarbonate solution (50 ml). The organic phase was dried and evaporated. The residue was a colourless oil (12.8 g, 59%). NMR ($CDCl_3$): 7.35 (s, C_6H_5); 5.73 (s, CL CH_2O); 5.10 (s, $OCH_2C_6H_5$); 4.33 (t, OCH_2CH_2N); 3.53 (t, OCH_2CH_2N); 2.92 (s, NCH_3).

3) To a stirred and ice-cooled suspension of 6-(α-carbonylphenylacetamido)-penicillanic acid α-benzyl monoester potassium salt (25.25 g, 50 mmole) in dry dimethylformamide (80 ml) was added dropwise chloromethyl-2-N-benzyloxycarbonylmethylaminoethylcarbonate (15.1 g, 50 mmole) in dry dimethylformamide (20 ml). After 16 hours stirring the reaction mixture was worked up in the usual way and the residue (26.8 g) was chromatographed on a silica gel column (400 g) using isopropylether-acetone (7:3) solvent system. A repeated chromatography from 14.3 g isolated substance in a similar manner gave the desired compound as a white foam (10.7 g, 27.8%) which was uniform according to TLC. IR (KBr): 1785—1765 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR ($CDCl_3$): 7.40 (s, C_6H_5); 7.25 (s, C_6H_5); 5.78 (s, OCH_2O); 5.65—5.40 (m, 5-H and 6-H); 5.16—5.10 (2s, $COOCH_2$ C_6H_5); 4.60 ($COCHC_6H_5$); 4.40 (d, 3-H); 4.25 (t. OCH_2CH_2N); 3.60 (t, OCH_2CH_2N); 2.95 (NCH_3); 1.50 (s, gem. CH_3).
Analysis: Calculated for $C_{37}H_{39}O_{11}N_3S$ (733.81); C 60.56; H 5.36; O 24.00; N 5.73; S 4.37. Found C 60.72; H 5.42; O 23.82; N 5.52; S 4.25.

4(The N-benzyloxycarbonyl α-benzylester derivative prepared above (1.5 g, 2 mmole) was hydrogenated in 50% dioxane (150 ml) over palladium-charcoal (3.0 g, Pd cont. 10%) at room temperature and normal pressure. After 3 hours the catalyst was filtered off and the pH of the filtrate was adjusted to 2.8 with 2N hydrochloric acid. The solution was evaporated to a diminished volume (ca 20 ml), extracted with ether several times, then the pH was adjusted to 4.8. After standing in refrigerator for several days a yellow-white powder precipitated, which was filtered off and washed with a few drops of cold water. IR (KBr): 2750—2530 (ammonium); 1780—1740 (β-lactam, ester and carbonate C=0); 1670 (amide C=0). Degree of hydrolysis: B3 = 23.8%; H3 =54.6%; R2 = 87.5%.

Example XIII.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl)-3-(2′-aminoethoxycarbonyloxymethyl) diester hydrochloride

1) According to the description given in example XI, 6-(α-carboxyphenylacetamido)-penicillanic acid α-(ethoxycarbonyloxymethyl) monoester sodium salt (4.09 g, 9.2 mmole) was treated with chloromethyl-(2-azidoethyl) carbonate (1.80 g, 10.0 mmole). Chromatography on silica gel (100 g) gave the diester in pure form

as a foam (1.41 g, 25%). IR ($CHCl_3$): 2150 (azido); 1780—1740 (β-lactam, ester and carbonate C=0) 1680 (amide C=0). NMR: 7.29 (d, 2 C_6H_5); 5.80 (s, OCH_2O); 5.80—5.40 (5-H and 6-H); 5.17 (s, $C_6H_5CH_2O$); 4.61 (d, C_6H_5CHCO); 4.45—4.20 (m, 3-H and $OCH_2CH_2N_3$); 3.49 (t. $OCH_2CH_2N_3$); 1.42 (s, gem. CH_3). Analysis: Calculated for $C_{28}H_{29}O_9N_5S$ (611.65; C 54.99; H 4.78; O 23.54; N 11.45; S 5.24. Found: C 55.16; H 4.94; O 23.42; N 11.36; S 5.17.

2) The azido-diester (920 mg, 1.5 mmole), prepared above, was hydrogenated in ethylacetate (55 ml) over palladium-charcoal (0.5 g, Pd. cont. 10%) at room temperature and normal pressure. After 7 hours the catalyst was filtered off, cold water (10 ml) was added to the filtrate, the pH was adjusted to 2.7 with 2N hydrochloric acid and the organic phase was separated and washed with water (2×5 ml). The combined water phase was extracted with isopropylether and then the water phase freeze-dried. The white, micro-crystalline residue was uniform according to TLC. IR(KBr): 3050 (ammonium); 1790—1775 (β-lactam, ester and carbonate C=0); 1685 (amide C=0); 1510 (ammonium). Degree of hydrolysis: B3 = 28.5%; H3 = 62.5%; R2 = 92.5%.

Example XIV.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid 3-(1'-cyclopentyloxycarbonyloxyethyl) ester sodium salt

1) A stream of dry chlorine (220 g, 3.14 mole) was passed through ethylchloroformate (450 g, 417 mole) at 25—35°C for 30 hours. During the reaction the mixture was irradiated with a 250 lamp (white light).

Fractional distillation of the product (the fractions were checked with GLC) gave one fraction containing pure (> 95%) α-chloroethylchloroformate (114 g, 25.6%).

2) The above obtained substance (50.0 g, 0.35 mole) was reacted with cyclopentanol (30.1 g, 0.35 mole) in the presence of pyridine (27.7 g, 0.35 mole) in the manner described in example Ia. After stirring for 19 hours, the reaction mixture was filtered and the filtrate was washed with 2N hydrochloric acid, saturated sodium bicarbonate solution, and water successively. After drying and evaporation the crude oil (60.3 g, 89%) was used directly in the next step.

3) α-Chloroethylcyclopentylcarbonate (2.89 g, 15.0 mmole) was added dropwise to a stirred and ice-cooled suspension of 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzylmonoester potassium salt (5.07 g, 10.0 mmole) in dry dimethylsulphoxide (10 ml). After stirring for 18 hours the mixture was worked up and chromatographed on silica gel (100 g) as in example XI. Pure 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzyl-3-(1'-cyclopentyloxycarbonyloxyethyl) diester (2.06 g, 33%) was isolated from one of the middle fractions of the eluate. IR ($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0) 1680 (amide C=0). NMR ($CDCL_3$): 7.28 (d, 2 C_6H_5); 6.78 (q, $OCH(CH_3)O$); 5.80—5.40 (m, 5-H and 6-H); 5.35—5.05 (m, $C_6H_5CH_2O$ and cyclopentyl); 4.60 (d, C_6H_5CHCO); 4.41 (d, 3-H); 2.00—1.70 (m, cyclopentyl); 1.60—1.10 (m, gem. CH_3 and $OCH(CH_3)O$). Analysis: Calculated for $C_{32}H_{36}O_9N_2S$ (624.73): C 61.53; H 5.81; O 23.05; N 4.48; S 5.13. Found: C 61.62; H 5.88; O 22.92; N 4.24; S 5.08.

3) The diester (1.87 g, 3.0 mmole) was hydrogenated and worked up in the same manner as in example IVa. The freeze-dried product (1.07 g, 64%) was pure according to TLC (butanone-pyridine-water-acetic acid system). IR (KBr): 1780—1740 (β-lactam, ester and carbonate C=0); 1680—1670 (amide C=0); 1610—1600 (carboxyl C=0). NMR (D_2O): 7.35 (s, C_6H_5); 6.71 (q, $OCH(CH_3)O$); 5.70—5.60 (m, 5-H and 6-H); 5.35—5.05 (m, cyclopentyl); 4.43 (s, 3-H); 2.00—1.70 (m, cyclopentyl); 1.50—1.10 (m, gem. CH_3 and $OCH(CH_3)O$). Analysis: Calculated for $C_{25}H_{29}O_9N_2SNa$ (556.57): N 5.03; S 5.76; Na 4.13. Found: N 4.92; S 5.58; Na 4.52. Degree of hydrolysis: B3 = 5.8%; H3 = 15.6%; R2 = 73.5%.

Example XV.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzyl-3-benzyloxycarbonyloxymethyl) diester

According to the description given in example XIV 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzylmonoester potassium salt (5.07 g, 10.0 mmole) was treated with chloromethylbenzylcarbonate (2.01 g, 10.0 mmole) to give, after chromatography on silica gel (100 g), the pure title compound (2.02 g, 32%) as a foam. IR ($CDCL_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amiede C=0). NMR ($CDCl_3$); 7.28 (d, 2 C_6H_5); 5.78 (s, OCH_2O); 5.80—5.40

(m, 5-H and 6-H); 5.18 (d, 2 $C_6H_5CH_2O$); 4.61 (d, C_6H_5CHCO); 4.42 (d, 3-H); 1.42 (s, gem. CH_3).
Analysis: Calculated for $C_{33}H_{32}O_9N_2S$ (632.70): C 62.65; H 5.10; O 22.76; N 4.43; S 5.07; Found: C 62.75; H 5.24; O 22.62; N 4.36; S 5.02. Degree of hydrolysis: B3 = < 1%; H3 = < 1%; R2 = 36.5%.

Example XVI.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid 3-(cis-2-methyl-1,3-dioxanyl-5-oxycarbonyloxymethyl) monoester sodium salt

Chloromethyl-5-(cis-2-methyl-1,3-dioxyanyl)-carbonate (1.68 g, 8.0 mmole) was reacted with 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzylmonoester potassium salt (4.05 g, 8.0 mmole) in the manner previously described. Chromatography gave the pure diester (1.44 g, 28%) as a foam. IR ($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1690—1680 (amide C=0). NMR ($CDCl_3$): 7.29 (d, 2 C_6H_5); 5.80 (s, OCH_2O); 5.80—5.40 (m, 5-H and 6-H); 5.19 (s, $C_6H_5CH_2O$); 4.69 (q, $OCH<(CH_2O)_2>CHCH_3$); 4.65—4.40 (m, 3.H, $OCH<(CH_2O)_2>CHCH_3$ and C_6H_5CHCO); 4.20—3.90

(m, $OCH<(CH_2O)_2>CHCH_3$);

1.60—1.10 (m, gem. CH_3 and $OCH<(CH_2O)_2>CHCH_3$).
Analysis: Calculated for $C_{31}H_{34}O_{11}N_2S$ (642.69): C 57.94; H 5.33; O 27.38; N 4.36; S 4.99. Found: C 57.82; H 5.24; O 27.42; N 4.30; S 4.72.

The diester (1.22 g, 1.9 mmole) was hydrogenated over palladium-charcoal in the usual manner to give, after freeze-drying, the title compound (0.72 g, 66%) as a white powder.
IR (KBr): 1780—1740 (β-lactam, ester and carbonate C=0) 1690—1680 (amide C=0); 1620—1600 (carboxyl C=0). NMR (D_2O): 7.35 (s, C_6H_5); 5.80—5.60 (m, OCH_2O, 5-H and 6-H); 4.70 (q, $OCH<(CH_2O)_2>CHCH_3$); 4.65—4.35 (m, 3-H, C_6H_5CHCO and $OCH<(CH_2O)_2>CHCH_3$); 4.20—3.90 (m, $OCH<(CH_2O)_2>CHCH_3$); 1.60—1.10 (m, gem. CH_3 and $OCH<(CH_2O)_2>CHCH_3$).
Analysis: Calculated for $C_{24}H_{27}O_{11}N_2SNa$ (574.54) N 4.88; S 5.58; Na 4.00. Found: N 4.66, S 5.42; Na 4.18. Degree of hydrolysis: B3 = 7.6%; H3 = 21.5%; R2 = 88.5%.

Example XVII.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α,3-(1'-ethoxycarbonyloxyethyl) diester

A suspension of sodium bicarbonate (15.1 g, 180 mmole) and 6-(α-carboxyphenylacetamido)-penicillanic acid 3-(1'-ethoxycarbonyloxyethyl) monoester sodium salt (15.5 g, 30 mmole) in 50% dioxane (30 ml) was added dropwise, with stirring and cooling in ice, α-chlorodiethylcarbonate (13.7 g, 90 mmole).

The reaction mixture was stirred for 64 hours, then it was filtered and to the filtrate was added chloroform (100 ml). The organic phase was separated and washed with water, saturated sodium bicarbonate solution and water successively. After evaporation the residue was kept under high vacuum (0.01 mmHg) for 12 hours to remove remaining dioxane and α-chlorodiethylcarbonate. Chromatography on silica gel (150 g) using the usual solvent system gave the title compound (1.50 g, 8.2%) in pure form isolated as a foam from one of the two main fractions of the eluate. The other main fraction contained 6-(phenylacetamido)-penicillanic acid 3-(1'-ethoxycarbonyloxyethyl) ester. IR ($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1690—1670 (amide C=0). NMR ($CDCl_3$): 7.39 (s, C_6H_5): 6.78 (q, $OCH(CH_3)O$); 5.80—5.40 (m, 5-H and 6-H); 4.60 (d, C_6H_5CHCO); 4.41 (d, 3-H); 4.20 (q, OCH_2CH_3); 1.60—1.10 (m, gem. CH_3, OCH_2CH_3 and $OCH(CH_3)O$).
Analysis: Calculated for $C_{27}H_{34}O_{12}N_2S$ (610.65): C 53.11; H 5.61; O 31.44; N 4.59; S 5.25. Found: C 53.25; H 5.72; O 31.28; N 4.46; S 5.15. Degree of hydrolysis: B3 = < 1%; H3 = 8.5%; R2 = 32.5%.

Example XVIII.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(1'-ethoxycarbonyloxyethyl) monoester sodium salt

a) *By method G*

1) To a stirred and ice-cooled suspension of phenylmalonic acid monobenzylester (40.5 g, 0.15 mole) and sodium bicarbonate (88.2 g, 1.05 mole) in

50% dioxane (150 ml) was added dropwise α-chlorodiethylcarbonate (68.6 g, 0.45 mole). Stirring was continued at room temperature for 64 hours.

The precipitate was filtered off, and to the filtrate was added chloroform (500 ml). The organic phase was separated and washed with water, saturated sodium bicarbonate solution, and water successively. After evaporation the residue (89.5 g) was kept under high vacuum (0.01 mm Hg) for 16 hours to remove remaining α-chlorodiethylcarbonate and dioxane. This residue (46.3 g) was chromatographed on a silica gel column (300 g), prepared in carbon tetrachloride. The substance was applied without dilution and was eluated with gradient technique, using dry chloroform as the second solvent.

As the second main fraction phenylmalonic acid benzyl-(1'-ethoxycarbonyloxyethyl) diester (9.7 g, 16.5%) was isolated as a colourless oil.

2) The diester (9.5 g, 24.6 mmole) was dissolved in ethyl acetate (100 ml) and hydrogenated at room temperature and normal pressure over palladium-charcoal (4.25 g, Pd cont. 5%) until one equivalent hydrogen had been absorbed. The catalyst was filtered off and the filtrate was evaporated to give phenylmalonic acid (1'-ethoxycarbonyloxyethyl) monoester (5.8 g, 80%) as a colourless syrup. IR (film): 3500—3200 (hydroxyl); 1760—1740 (ester and carbonate C=0); 1690 (carboxyl C=0). NMR ($CDCl_3$): 10.20 (s, COOH); 7.32 (s, C_6H_5); 6.78 (q, $OCH(CH_3)O$); 4.65 (s, C_6H_5CHCO); 4.12 (q, OCH_2CH_3); 1.60—1.10 (m, OCH_2CH_3 and $OCH(CH_3)O$).

3) Phenylmalonic acid (1'-ethoxycarbonyloxyethyl) monoester chloride (3.15 g, 10 mmole), prepared from the corresponding acid (2.96 g, 10 mmole) in the manner previously outlined, was used to acylate sodium 6-aminopenicillinate using the method described in example Ib. The freeze-dried product (3.10 g, 60%) showed a main spot on TLC (butanone-pyridine-water-acetic acid system), besides a minor quantity of disodium 6-(α-carboxyphenylacetamido)-penicillinate. IR (KBr); 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0); 1610 (carboxyl C=0). NMR (D_2O): 7.40 (s, C_6H_5); 6.70 (q, $OCH(CH_3)O$); 5.70—5.60 (m, 5-H and 6-H); 4.30 (s, 3-H); 4.12 (q, OCH_2CH_3); 1.60—1.10 (m, OCH_2CH_3, $OCH(CH_3)O$ and gem. CH_3).
Analysis: Calculated for $C_{22}H_{25}O_9N_2SNa$ (516.51): N 5.42; S 6.21; Na 4.45. Found: N 5.28; S 6.12; Na 4.58. Degree of hydrolysis: B3 = 4.8%; H3 = 13.8%; R2 = 85.3%.

b) *By method H*

1) A suspension of 6-(α-carboxyphenylacetamido)-penicillanic acid 3-benzylhydrylmonoester sodium salt (17.0 g, 30 mmole), (prepared by acylating 6-aminiopenicillanic acid 3-benzylhydrylester p-toluenesulphonate with phenylmalonic acid monochloride) and sodium bicarbonate (15.1 g, 180 mmole) in 50% dioxane (30 ml) was treated with α-chlorodiethylcarbonate (13.7 g, 90 mmole) in a similar manner as described in example XVII. After working up and chromatography on silica gel pure 6-(α-carboxyphenylacetamido)-penicillanic acid α-(1'-ethoxycarbonyloxyethyl)-3-benzhydryl diester (1.74 g, 8.8%) was isolated from the second main fraction of the eluate.
IR ($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR ($CDCl_3$): 7.38 (d, 3 C_6H_5); 6.95 (s, $(C_6H_5)_2CH$); 6.78 (q, $OCH(CH_3)O$); 5.80—5.40 (m, 5-H and 6-H); 4.66 (d, C_6H_5CHCO); 4.51 (d, 3.H); 4.20 (q, OCH_2CH_3); 1.60—1.10 (m, OCH_2CH_3, $OCH(CH_3)O$ and gem. CH_3). Analysis: Calculated for $C_{35}H_{36}O_9N_2S$ (660.76); C 63.62; H 5.49; O 21.79; N 4.24; S 4.85. Found: C 63.78; H 5.62; O 21.68; N 4.16; S 4.32.

2) The diester (1.65 g, 2.5 mmole) was hydrogenated over palladium-charcoal using the same method and working up procedure as described in example Ia. The product (0.92 g, 71%) was by its spectral, analytical and hydrolysis data identical with, but purer than, the substance prepared by method E.

Example XIX.

Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-(5'-indanyloxycarbonyloxymethyl) monoester sodium salt

Sodium 6-aminopenicillinate in 50% acetone solution was acylated with phenylmalonic acid (5'-indanyloxycarbonyloxymethyl) monoester chloride (1.17 g, 3.0 mmole) using the same method and working up procedure as in example Ib. The freeze-dried product (1.05 g, 59%) showed one main spot on TLC (butanone-pyridine-water-acetic acid system) besides a minor quantity of disodium 6-(α-carboxyphenylacetamido)-penicillinate.

IR (KBr): 1780—1740 (β-lactam, ester and carbonate C=0); 1690—1680 amide C=0); 1610—1600 (carboxyl C=0). NMR (D_2O): 7.40—6.90 (m, C_6H_5 and indanyl-H); 5.80—5.50 (m, OCH_2O, 5-H and 6-H); 4.30 (s, 3-H); 2.89 (t, indanyl-H); 2.30—1.90 (m, indanyl-H); 1.50 (s, gem. CH_3). Analysis: Calculated for $C_{28}H_{27}O_9N_2SNa$ (590.59): N 4.74; S 5.43; Na 3.89. Found: N 4.66: S 5.32; Na 4.12. Degree of hydrolysis: B3 = 17.2%; H3 = 35.4%; R2 = 83.6%.

Example XX.
Pharmaceutical formulations

For preparation of tablets the following compositions were made.

a) Sodium 6-(α-(ethoxycarbonyloxymethoxy)carbonyl-phenylacetamido)penicillanate	350 mg
Starch	100 mg
Magnesium stearate	10 mg
b) Sodium 6-(α-(ethoxycarbonyloxymethoxy)carbonyl-3-thienylacetamido)penicillanate	400 mg
Starch	100 mg
Magnesium stearate	10 mg
c) Ethoxycarbonyloxymethyl 6-(α-(ethoxycarbonyloxy-methoxy)carbonylphenyl-acetamido)penicillanate	500 mg
Calcium carbonate	100 mg
Magnesium stearate	10 mg
d) 1'-Ethoxycarbonyloxymethyl 6-(α-carboxyphenyl-acetamido)penicillanate sodium salt	400 mg
Lactose	100 mg
Magnesium stearate	10 mg
e) Sodium 6-(α-(1'-ethoxycarbonyloxyethoxy)carbonyl-phenylacetamido)penicillanate	400 mg
Microcrystalline cellulose (Avice)®	100 mg
Magnesium stearate	10 mg

For filling in capsules the following formulations were made:

(f) Sodium 6-(α-(ethoxycarbonyloxymethoxy)carbonyl-phenylacetamido)penicillanate	350 mg
Magnesium stearate	5 mg

For oral suspensions the following formulations were prepared:

(g) Sodium 6-(α-(1'-ethoxycarbonyloxyethoxy)carbonyl-phenylacetamido)penicillanate	35 g
Sodium benzoate	0.48 g
Sodium chloride	0.75 g
Flavouring agents	4.7 g
Aerosil®	0.3 g
Antifoam®	0.0375 g
Alkali salts of polysaccharide sulphates	4.0 g
Sodium saccharinate	0.4 g
Sorbitol	ad 100 g

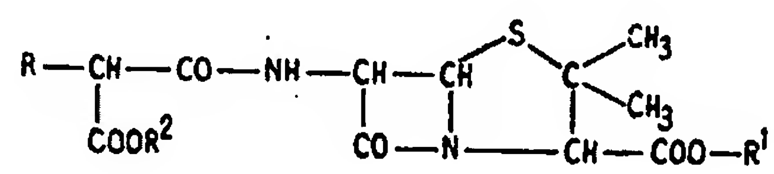
Example XXI.
Preparation of 6-(α-carboxyphenylacetamido)-penicillanic acid α-benzyl-3-(2-furfuryloxycarbonyloxymethyl)diester

Using the same method as in example XIV and XV, 8-(α-carboxyphenyl-acetamido)-penicillanic acid α-benzylmonoester potassium salt (5.07 g, 10.0 mmole) was treated with chloromethyl-(2-furfuryl)-carbonate (1.91 g, 10.0 mmole). Chromatography gave the pure diester (1.68 g, 27%) as a foam. IR ($CHCl_3$): 1780—1740 (β-lactam, ester and carbonate C=0); 1680 (amide C=0). NMR ($CDCl_3$): 7.50—7.30 (m, C_6H_5 and C_4H_3O); 6.55—6.35 (m, C_4H_3O); 5.78 (s, OCH_2O); 5.80—5.40 (m, 5-H and 6-H); 5.18 (s, $C_6H_5CH_2O$ and $C_4H_3OCH_2O$);

Analysis: Calculated for $C_{31}H_{30}O_{10}N_2S$ (622.66): C 59.80; H 4.86; O 25.70; N 4.50; S 5.15. Found: C 60.02; H 4.78; O 25.54; N 4.32; S 5.04. Degree of hydrolysis: $B_3 = < 1\%$; $H3 = < 1\%$; $R2 = 41.5\%$.

WHAT WE CLAIM IS:—

1. A compound of the general formula



I

and pharmaceutically acceptable salts thereof, in which
R is phenyl, thienyl or furyl group and
R^1 is hydrogen or a

$$-\overset{\overset{\displaystyle R^3}{|}}{C}H-O-\overset{\overset{\displaystyle O}{\|}}{C}-O-R^4 \text{ group and}$$

R^2 is a

$$-\overset{\overset{\displaystyle R^3}{|}}{C}H-O-\overset{\overset{\displaystyle O}{\|}}{C}-O-R^4$$

group or hydrogen or an alkyl group of 1 to 8 carbon atoms, an aryl group or an aralkyl group,
R^3 is hydrogen or a methyl group;
R^4 is an alkyl, alkenyl or alkynyl group of up to 8 carbon atoms, a cycloalkyl group of 3 to 7 carbon atoms or a phenyl, benzyl, indanyl, thienyl, furyl, furfuryl, pyridyl, pyridylmethyl or 2-methyl-1,3-dioxanyl group, the said groups being unsubstituted or substituted with one or more amino, substituted amino, halogeno or nitro radicals;
provided that R^2 is

$$-\overset{\overset{\displaystyle R^3}{|}}{C}H-O-\overset{\overset{\displaystyle O}{\|}}{C}-O-R^4 \text{ when } R^1 \text{ is a hydrogen atom.}$$

2. A compound according to claim 1 wherein the substituted amino group is methylamino, diethylamino or acetamido.

3. A compound according to claim 1 or 2 wherein R^1 is hydrogen.

4. A compound according to claim 1 or 2 wherein R^2 is hydrogen.

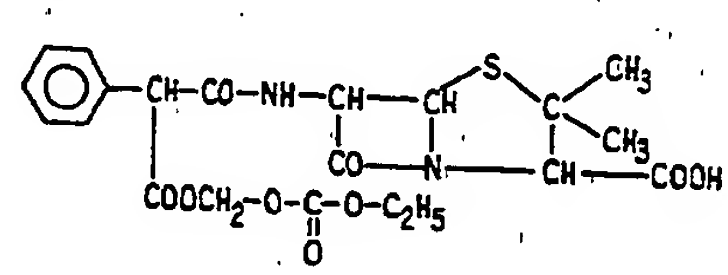
5. A compound according to claim 1 wherein R^2 is hydrogen, alkyl, benzyl, phenyl, 5-indanyl, alkoxycarbonyloxymethyl, 1′-alkoxycarbonyloxyethyl, phenoxycarbonyloxymethyl, 5-indanyloxycarbonyloxymethyl, 1′-phenoxycarbonyloxy-ethyl, 1′-(5-indanyloxy) carbonyloxy-ethyl, and R^1 is alkoxycarbonyloxymethyl, 1′-alkoxycarbonyloxyethyl, phenoxycarbonyloxy-methyl, 5-indanyloxycarbonyloxymethyl, 1′-phenoxycarbonyloxy-ethyl, or 1′-(5-indanyloxy)carbonyloxy-ethyl, the aforesaid alkoxy groups containing 1—8 carbon atoms.

6. A compound according to claim 1 wherein R^1 is hydrogen and R^2 is alkoxy-carbonyloxymethyl, 1′-alkoxycarbonyloxy-ethyl, phenoxycarbonyloxy-methyl, 5-indanyloxycarbonyloxy-methyl, 1′-phenoxycarbonyloxy-ethyl, or 1′-(5-indanyloxy)carbonyloxy-ethyl, the aforesaid alkoxy group containing 1—8 carbon atoms.

7. A compound according to claim 5 or 6 wherein the alkoxycarbonyloxy groups in R^1 and/or R^2 are substituted by amino, methylamino or di-alkylamino groups.

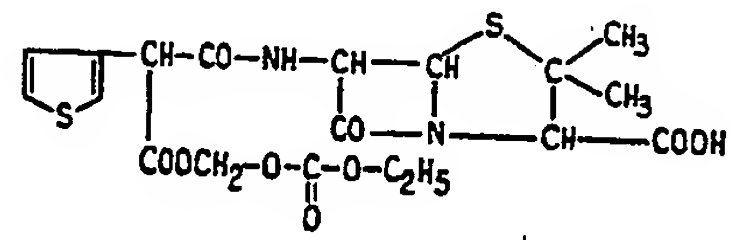
8. A compound according to any one of the preceeding claims wherein R^4 is tert-amyl, β-aminoethyl, ethyl, cyclopentyl, β-methylaminoethyl, β-acetamido-ethyl, β-chloroacetamido-ethyl, β-thioacetamidoethyl, thienyl, benzyl, allyl, indanyl, isopropyl, methyl, 3,3-dimethylbutyl, pyridylmethyl, furfuryl, phenyl or chlorophenyl.

9. A compound of formula



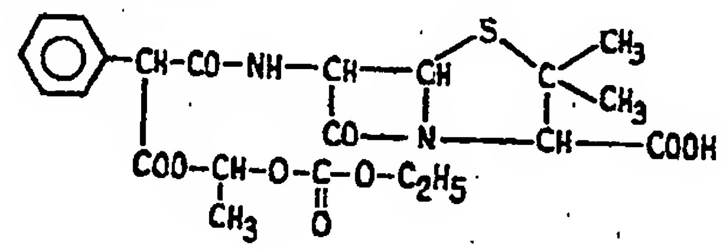
or a therapeutically acceptable salt thereof.

10. A compound of formula



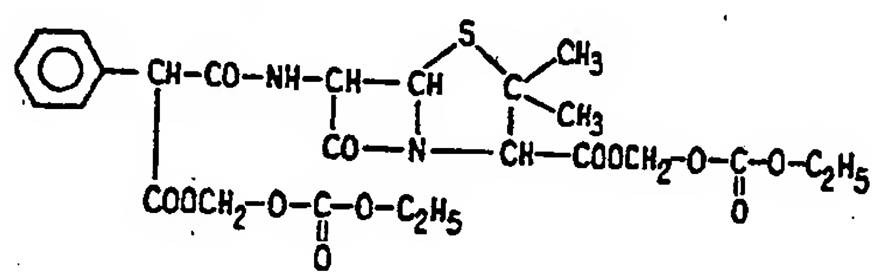
or a therapeutically acceptable salt thereof.

11. A compound of formula



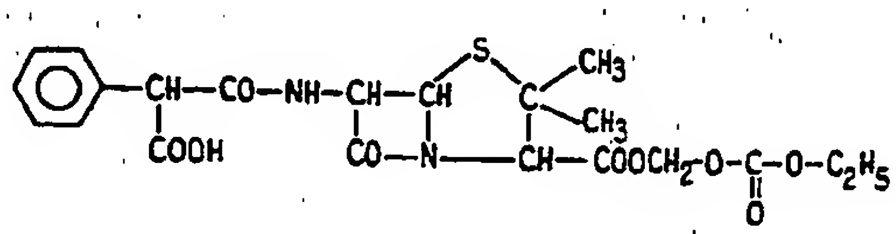
or a therapeutically acceptable salt thereof.

12. A compound of formula



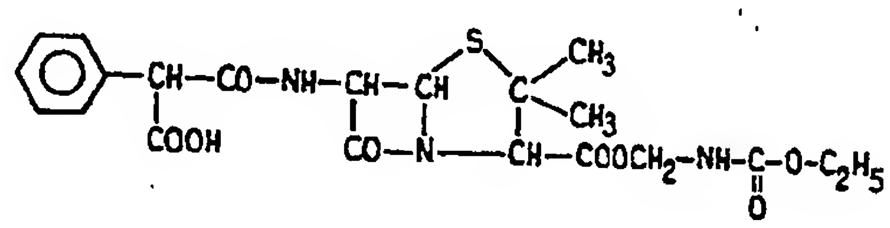
or a therapeutically acceptable salt thereof.

13. A compound of formula



or a therapeutically acceptable salt thereof.

14. A compound of formula



or a therapeutically acceptable salt thereof.

15. A compound according to claim 1 hereinbefore specifically mentioned.

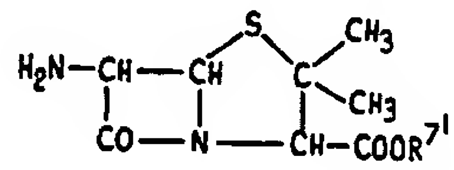
16. A compound according to any one of the preceding claims having at least one asymmetric centre in the form of a substantially pure stereo isomer.

17. A compound according to any one of the preceding claims in the form of a monoester sodium salt.

18. A process for the preparation of a compound as defined in claim 1 where R^1 is $-CH(R^3)OCOOR^4$ which comprises reacting a compound of the formula

$$\begin{array}{l} R{-}\underset{|}{CH}{-}CO{\sim}7 \\ \quad COOR^{2'} \end{array} \qquad \text{III}$$

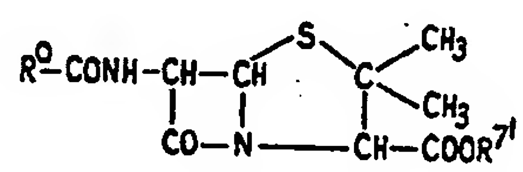
with a compound of the formula



IV

in which R is as defined in claim 1, $R^{2'}$ is R^2, as defined in claim 1, or, when R^2 is hydrogen or when R^2 contains amino or substituted amino groups is a protected derivative of R^2, —CO—Z is a reactive group capable of reacting with an amino group to form an amide, and $R^{7'}$ is as defined in this claim for R^1, or, when R^1 contains an amino or protected amino group, is a protected R^1 group; and, if necessary, then removing any amino, substituted amino or carboxy protecting groups.

19. A process for the preparation of a compound as defined in claim 1 where R^1 is —$CH(R^3)OCOOR^4$ which comprises reacting an ester of a natural or biosynthetic penicillin of the formula



VIII

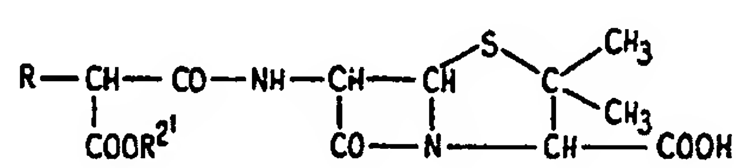
wherein R^o—CO— represents the acyl group of the side chain of the natural or biosynthetic penicillin, with a phosphorus halide in an inert solvent; reacting the resulting imino halide with a lower alcohol, and reacting the resulting imino ether with a compound of the formula

$$\begin{array}{l} R—CH—CO—Z \\ \qquad | \\ \qquad COOR^{2'} \end{array}$$

III

in which R, $R^{2'}$, $R^{7'}$ and COZ are as defined in claim 18 and then removing any amino, substituted amino or carboxy protecting groups.

20. A process for the preparation of a compound as defined in claim 1 where R^1 is —$CH(R^3)OCOOR^4$ which comprises reacting a compound of the formula



IX

with a compound of the formula:

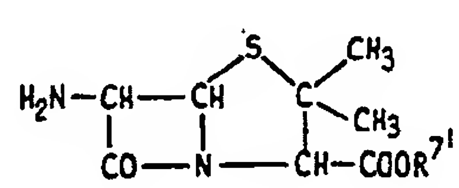
$$R^{7'} — Y$$

where R, $R^{2'}$ and $R^{7'}$ are as defined in claim 18 and Y is halogen or other group that reacts to form an ester link; and then, if necessary, removing any amino, substituted amino or carboxy protecting groups.

21. A process for the preparation of a compound as defined in claim 1 where R^1 is —$CH(R^3)OCOOR^4$ which comprises reacting a compound of the formula

$$\begin{array}{l} R—C{=}C{=}O \\ \qquad | \\ \qquad COOR^{2'} \end{array}$$

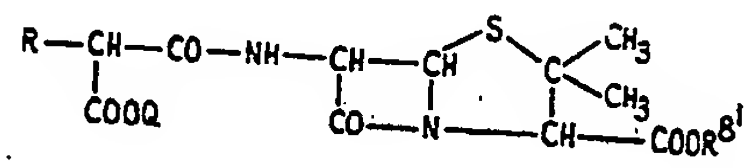
with a compound of the formula



IV

where R is as defined in claim 1 and $R^{2\prime}$ and $R^{7\prime}$ are as defined in claim 18; and then, if necessary, removing any amino, substituted amino or carboxy protecting groups.

22. A process for the preparation of a compound as defined in claim 1 except those where R^2 and/or R^1 is hydrogen which comprises reacting a compound of the formula



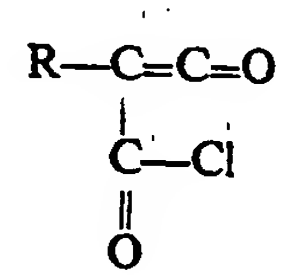
X

with a compound of the formula

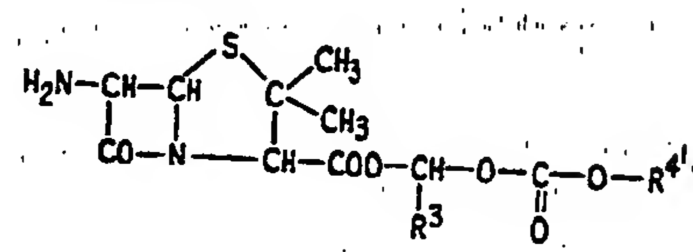
$$R^{6\prime} — Y$$

where $R^{6\prime}$ and $R^{8\prime}$ are as defined for R^2 and R^1 respectively in claim 1 with the proviso that neither can represent hydrogen, or, where $R^{6\prime}$ and/or $R^{8\prime}$ contains an amino or substituted amino group, is a protected $R^{6\prime}$ or protected $R^{8\prime}$ group, R is as defined in claim 1, Q is H or a cation, and Y represents halogen or ether group which reacts to form an ester link; and then, if necessary removing any amino or substituted amino protecting groups.

23. A process for the preparation of a compound as defined in claim 1 where R^2 is hydrogen and R^1 is —$CH(R^3)$—$OCOOR^4$ which comprises reacting a compound of the formula



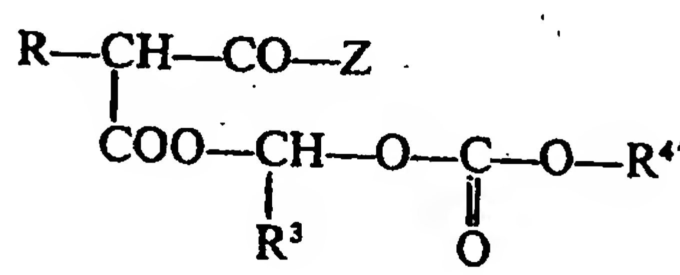
with a compound of the formula



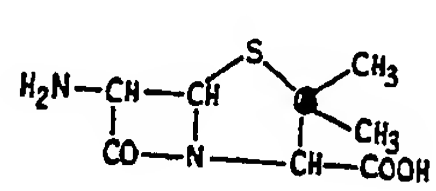
XIV

where $R^{4\prime}$ represents R^4 as defined in claim 1 or, when it contains an amino or substituted amino group, is a protected $R^{4\prime}$ group and R is as defined in claim 1; and then, if necessary, removing any amino or substituted amino protecting groups.

24. A process for the preparation of a compound as defined in claim 1 where R^1 is hydrogen and R^2 is —$CH(R^3)$—$OCOOR^4$ which comprises reacting a compound of the formula



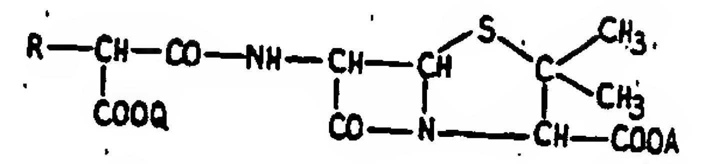
with a compound of the formula



where $R^{4\prime}$, R and R^3 are as defined in claim 1 and is as defined in claim 23 and COZ is a reactive group capable of reacting with an amino groups to form an amide; and then, if necessary, removing any amino or substituted amino protecting groups.

25. A process for the preparation of a compound as defined in claim 1 where

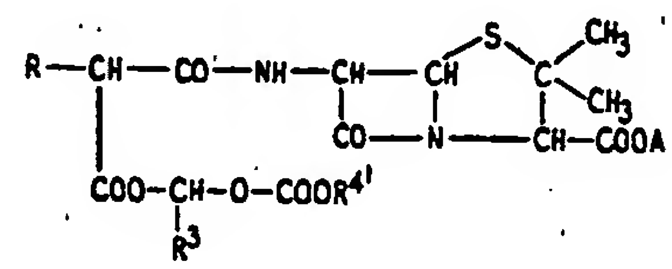
R^1 is hydrogen and R^2 is —$CH(R^3)OCOOR^4$ which comprises reacting a compound of the formula



with a compound of the formula

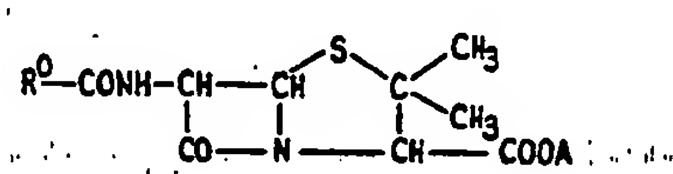
$$Y-\underset{|}{\overset{R^3}{CH}}-O-\overset{O}{\overset{\|}{C}}-O-R^{4'}$$

to form a compound of the formula

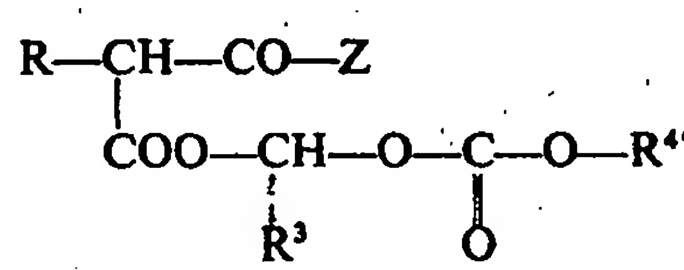


and then converting COOA to COOH, where $R^{4'}$ is as defined in claim 23, Y is halogeno or other group that reacts to form an ester link, R and R^3 are as defined in claim 1, Q is as defined in claim 22, and A is a carboxy protecting group and, if necessary, removing any amino or substituted amino protecting groups before, during or after removal of the carboxy protecting group.

26. A proces for the preparation of a compound as defined in claim 1 where R^1 is hydrogen and R^2 is —$CH(R^3)OCOOR^4$ which comprises reacting a natural or biosynthetic penicillin of the formula

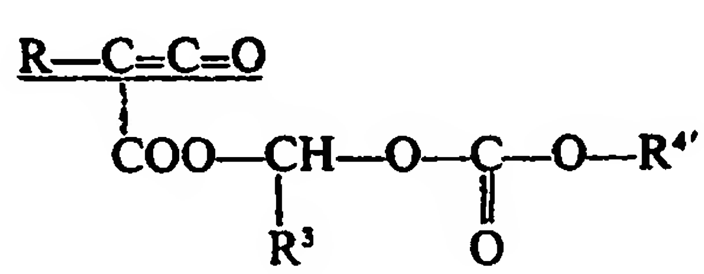


wherein R° —CO— represents the acyl group of the side chain of the natural or biosynthetic penicillin, with phosphorus halide in an inert solvent; reacting the resulting imino halide with a lower alcohol and reacting the resulting imino ether with a compound of the formula

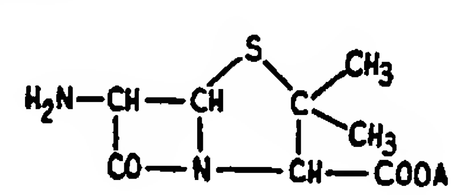


wherein $R^{4'}$ is as defined in claim 23, COZ is as defined in claim 24, R and R^3 are as defined in claim 1, and A is as defined in claim 25 and then removing the carboxy protecting group and, if necessary, any amino or substituted amino protecting groups.

27. A process for the preparation of a compound as defined in claim 1 where R^1 is hydrogen and R^2 is —$CH(R^3)OCOOR^4$ which comprises reacting a compound of the formula

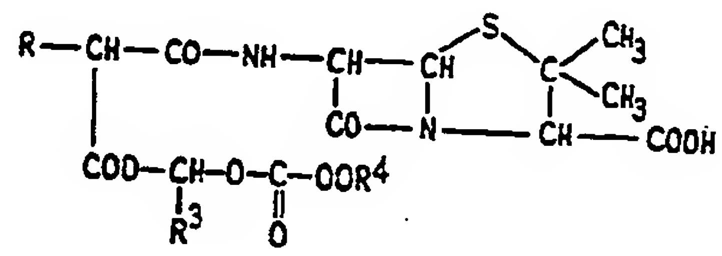


with a compound of the formula

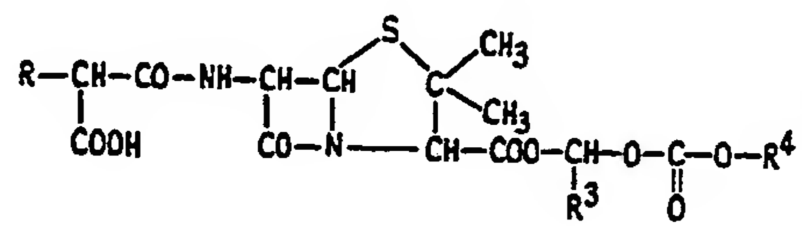


wherein $R^{4'}$ is as defined in claim 23, R and R^3 are as defined in claim 1, and A is as defined in claim 25 and then removing the carboxy protecting group and, if necessary, any amino or substituted amino protecting groups.

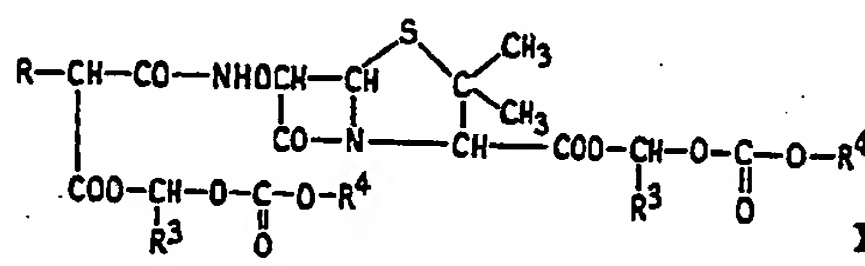
28. A process for the preparation of a mixture of compounds of the formula:



XVIIIA

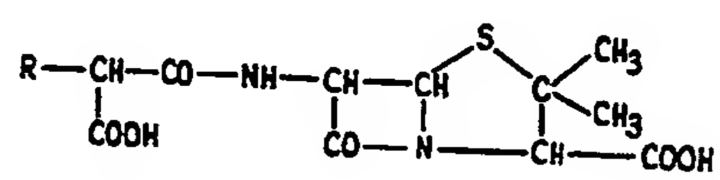


XXVA and



XXVIA

which comprises reacting a compound of the formula



with a compound of the formula

$$Y\text{—}\underset{}{\overset{R^3}{\overset{|}{C}}}H\text{—}O\text{—}\overset{O}{\overset{\|}{C}}\text{—}O\text{—}R^{4'}$$

where $R^{4'}$ is as defined in claim 23, R, R^3 and R^4 are as defined in claim 1 and Y is as defined in claim 24 and then, if necessary, removing any amino or substituted amino protecting groups.

29. A process according to claim 28 wherein at least one compound of formula XVIIIA, XXVA or XXVIA is separated from the mixture by a known method.

30. A process according to claim 20, 28 or 29 wherein the carboxy penicillin is reacted in the form of a tetraalkylammonium salt.

31. A process according to claim 30 wherein the salt is tetrabutyl ammonium.

32. A process according to claim 30 or 31 wherein the reaction is carried out in chloroform, methylene, chloride or acetone.

33. A process according to any one of claims 18—32 wherein the compound is converted to a pharmaceutically acceptable salt by reaction with a pharmaceutically acceptable acid or base.

34. A process according to any one of claims 18—33 wherein the compound or salt has at least one asymmetric centre and is resolved into its stereoisomers.

35. A process according to any one of claims 18—34 substantially as hereinbefore described.

36. A compound or salt obtained by a process according to any one of claims 18—35.

37. A pharmaceutical composition comprising a compound or salt according to any one of claims 1—17 or 36 together with a pharmaceutically acceptable carrier and/or adjuvant.

38. A composition according to claim 37 substantially as hereinbefore described.

39. A method of combatting infection in animals excluding man which comprises administering to the animal a compound or salt according to any one of claims 1—17 or 36 or a composition according to claim 37 or 38.

J. A. KEMP & CO.,
Chartered Patent Agents,
14 South Square,
Gray's Inn, London, W.C.1.

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